



Missouri
Department of
Natural Resources

Biological Assessment and Fine Sediment Study Report

Tributaries of Mill Creek and Mineral Fork, Part II: Pond Creek, Shibboleth Branch, and Tributary of Mineral Fork, Washington County

Fall 2010 – Spring 2011

Prepared for:

Missouri Department of Natural Resources
Division of Environmental Quality
Water Protection Program
Water Pollution Control Branch

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ATTACHMENTS

- Appendix A Macroinvertebrate Database Bench Sheet Report for Mill Creek
 Tributaries and Trib. Mineral Fork Stations, Fall 2010-Spring 2011

- Appendix B Fine Sediment Percent Coverage Statistics: Kruskal-Wallis One Way
 ANOVA on Ranks with Dunn's Test Multiple Comparisons of Test
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1.0 Introduction

This project is a continuation of the Biological Assessment and Fine Sediment Study Report: Tributaries of Mill Creek and Mineral Fork, Washington County, Fall 2008-Spring 2009 – Fall 2009 (Missouri Department of Natural Resources, **MDNR** 2009a). The earlier study included biological assessments and fine sediment studies at Pond Creek #2 and #1 (Water Body Identification (**WBIDs**) 2128 and 2127, respectively) as well as Shibboleth Branch #3 and #1 (WBIDs 2120 and 2119, respectively). Previous results will be compared with the current study when appropriate. This study includes the same stations on Pond Creek and Shibboleth Branch, as well as two new stations on Tributary (**Trib.**) of Mineral Fork (WBID 2115).

These tributaries of Mill Creek and Mineral Fork are located in southeastern Missouri within the Ozark/Meramec Ecological Drainage Unit (**EDU**; Table 1; Figure 1). The streams ultimately drain into Big River approximately 60 miles southwest of St. Louis, Missouri. The tributaries are located in Washington County (Table 1; Figure 2), whereas the similar-size control streams are located in Crawford, Dent, and Iron counties, Missouri (Table 1; MDNR 2009a). Control stream data were collected in the earlier study (MDNR 2009) and are applied in this study where appropriate.

Most of the tributaries included in this study are listed as class “C” or “P” streams in Missouri’s Water Quality Standards (Table 1; MDNR 2010e). Class “C” streams may cease flow in dry periods, but maintain permanent pools which support aquatic life. Class “P” streams maintain permanent flow even during drought periods. Because the streams included in this study are generally very small, one unclassified (shown as class “U”) station on Courtois Creek was chosen as a control segment.

Most of the streams in this study have beneficial use designations for livestock and wildlife watering (**LWW**); protection of warm water aquatic life and human health-fish consumption (**AQL**); and whole body contact (**WBC**), category B (MDNR 2010e). The WBC “Category B” applies to waters designated for whole body contact recreation not contained within category A. Category A is defined as:

Those water segments that have been established by the property owner as public swimming areas allowing full and free access by the public for swimming purposes and waters with existing whole body contact recreational use(s). Examples of this category include, but are not limited to, public swimming beaches and property where whole-body contact recreational activity is open to and accessible by the public through law or written permission of the landowner (MDNR 2010e).

An example of a category B stream may be one that is used for swimming, but is not designated specifically for such use.

1.1 Justification

The watersheds of Mill Creek and Mineral Fork in Washington County have been extensively mined for barium. In the fall of 2005 and spring of 2006, the Environmental Services Program (**ESP**), Water Quality Monitoring Section (**WQMS**) conducted biological assessments on Mill Creek and Mineral Fork, Washington County (MDNR 2007a, 2007b). Mill Creek contained high dissolved barium concentrations, apparently either from runoff within the watershed or from instream natural background occurrences. Mineral Fork had relatively high concentrations of dissolved barium with a continuous low level of chloride. The presence of chloride can be an indicator of mining activity and/or wastewater influence. The tributaries of these streams were recommended for study as potential contributors of mine-related material.

Heavy metals associated with mine related activity have been found in aquatic organisms. Crayfish and other aquatic macroinvertebrates were found to accumulate elevated concentrations of metals at mine related streams in southeast Missouri (Besser et al. 1987, 2007; Poulton et al. 2009; Allert et al. 2008, 2009, 2011). Macroinvertebrate communities appear to be negatively affected by mining activities where elevated concentrations of metals are found in sediment pore water (Besser et al. 2007, 2009a, 2009b; Brumbaugh et al. 2007; Poulton et al. 2009; Allert et al. 2008, 2011). Heavy metals have also been found in fish of Mill Creek in an earlier study (Czarnecki and Trial 1997). Metals such as copper, iron, lead, and zinc have been detected in aquatic fauna in areas of Big River (Czarnecki et al. 1997; Missouri Department of Conservation (**MDC**) 1997, 2006). Continued monitoring of heavy metals in fish tissue has led to present consumption advisories in the Big River watershed (Missouri Department of Health and Senior Services (**MDHSS**) 2012). Heavy metals such as zinc have been found specifically in the fine sediments of Pond Creek, whereas cadmium, lead, and zinc were found in fine sediments of Shibboleth Branch in the earlier tributaries study (MDNR 2009a). Concentrations of these metals in the sediment were above probable effects concentrations (**PECs**; MacDonald et al. 2000).

Historically, mine waste sedimentation has been responsible for covering aquatic habitats making them uninhabitable for some invertebrates (Ryck 1974; MDC 1997, 2006). Fine sediments and silt clog the interstitial voids between the larger particles in the substrate and can have destructive effects on invertebrates and fish communities (Chutter 1969; Murphy et al. 1981; Berkman and Rabeni 1987; Smale et al. 1995). Damage to some aquatic habitats and the potential for serious damage to several streams existed due to past lead and barite mining activity (MDC 1997, 2006). In 1975, the collapse of a barite tailings pond released a significant amount of metals-laden fine sediment into Shibboleth Creek, a tributary of Mill Creek (Duchrow 1978). Fine sediment coverage was considered a possible contributor to the consistent impairment of Pond Creek #2 and Shibboleth Branch #3 in the earlier MDNR (2009a) study.

Pond Creek and Shibboleth Branch are tributaries of Mill Creek and were placed on the Clean Water Act Section 303(d) list of impaired waters in 1998 (USEPA 2009; MDNR

2009b). Approximately one mile of Pond Creek (WBID 2128; incorrectly labeled *Tributary to* Pond Creek) was placed on the 303(d) list for inorganic sediment with a potential source being a barite tailings pond (MDNR 2010e). Approximately three miles of Shibboleth Branch (WBID 2120; mislabeled *Creek*) were initially placed on the list for inorganic sediment, and in 2010 for sediment lead and zinc, potentially from a mill source (EPA 2009; MDNR 2009b). Trib. Mineral Fork is not listed; however, it flows adjacent to an abandoned smelter and drains a barite tailings pond once known as the Dorlac Lake. The dam has a high hazard rating with an unsafe designation (MDC 1997).

This study was requested by the MDNR, Water Protection Program (**WPP**), Water Pollution Control Branch (**WPCB**). The 2010-2011 biological assessment and fine sediment study was conducted by the Division of Environmental Quality (**DEQ**), ESP, WQMS and Chemical Analysis Section (**CAS**).

This study includes stream habitat assessments, biological assessments, dissolved metals analysis in surface and pore water using instream diffusion samplers known as “peepers” (Serbst et al. 2003; Brumbaugh et al. 2002, 2007), and fine sediment relative percent coverage and metals character.

1.2 Objectives

- Assess the quality of stream habitat.
- Assess the “protection of aquatic life” designated use status using the macroinvertebrate community.
- Assess physicochemical water quality.
- Analyze surface water dissolved metals concentrations.
- Analyze substrate pore water metals concentrations.
- Determine the relative coverage of fine sediment per area and identify the metals character of sediment.

1.3 Null Hypotheses

1. Stream habitat quality will be similar between test and control tributaries.
2. Biological metrics and Macroinvertebrate Stream Condition Index (**MSCI**) scores will be similar between test and control streams as well as wadeable/perennial stream biological criteria.
3. Physicochemical water quality will be similar among stations, and parameters will meet the Water Quality Standards (**WQS**) of Missouri (MDNR 2010e).

4. The relative coverage and metals character of fine sediment in test streams will be similar to the control streams, and metals concentrations will be below threshold levels.

2.0 Methods

Kenneth B. Lister, Brandy S. Bergthold, and others of the ESP, WQMS staff conducted this study. Methods and study timing are outlined in this section. The study area and station descriptions, EDUs, and land uses are identified. Stream habitat assessment procedures are discussed. Biological assessment procedures, which include macroinvertebrate community and physicochemical water collection with analyses, are discussed in this section. Instream diffusion samplers (peepers) were used and methods for their use are discussed. Fine sediment relative percent estimation and characterization are outlined in this section.

2.1 Study Timing

Sampling was conducted in the fall of 2010 and the spring of 2011. Fall macroinvertebrate and water quality samples were collected on September 21, 2010, at Trib. Mineral Fork stations and Shibboleth Branch #1. Pond Creek stations and Shibboleth Branch #3 were sampled on September 22, 2010. Habitat assessments and the fine sediment studies were conducted at Trib. Mineral Fork stations on August 31, 2010. A fine sediment sample was collected at Trib. Mineral Fork #2 on September 21, 2010.

Peepers were deployed for 23 or 24 days. Peepers were deployed on Trib. Mineral Fork stations and Shibboleth Branch #1 on September 21, 2010. One or two peepers were deployed at Pond Creek and Shibboleth Branch #3 on September 22, 2010. Peepers were retrieved from all stations on October 14, 2010, with the exception of Pond Creek #2 where both peepers were missing.

Spring macroinvertebrates and water quality samples were collected on March 23, 2011, at Pond Creek stations and Shibboleth Branch #1. Trib. Mineral Fork and Shibboleth Branch #3 samples were collected on March 24, 2011.

2.2 Study Area, Station Locations and Descriptions

The study area and station locations for the 2010-2011 tributaries project were in the Ozark/Meramec EDU see 2.2.1; Table 1; Figure 1). Two stations were allocated for each of the three tributaries in this project (Table 1; Figure 2). Five WBIDs codes were examined and stations were positioned to observe potential influences. Pond Creek stations #2 (WBID 2128) and #1 (WBID 2127); Shibboleth Branch stations #2 (WBID 2120) and #1 (WBID 2119); and Trib. Mineral Fork stations #2 and #1 (WBID 2115) were numbered from upstream (high) to downstream (low). The control streams from the 2009 MDNR study were used for comparisons of stream habitat, dissolved metals in surface water and pore water, and fine sediment relative quantity and character.

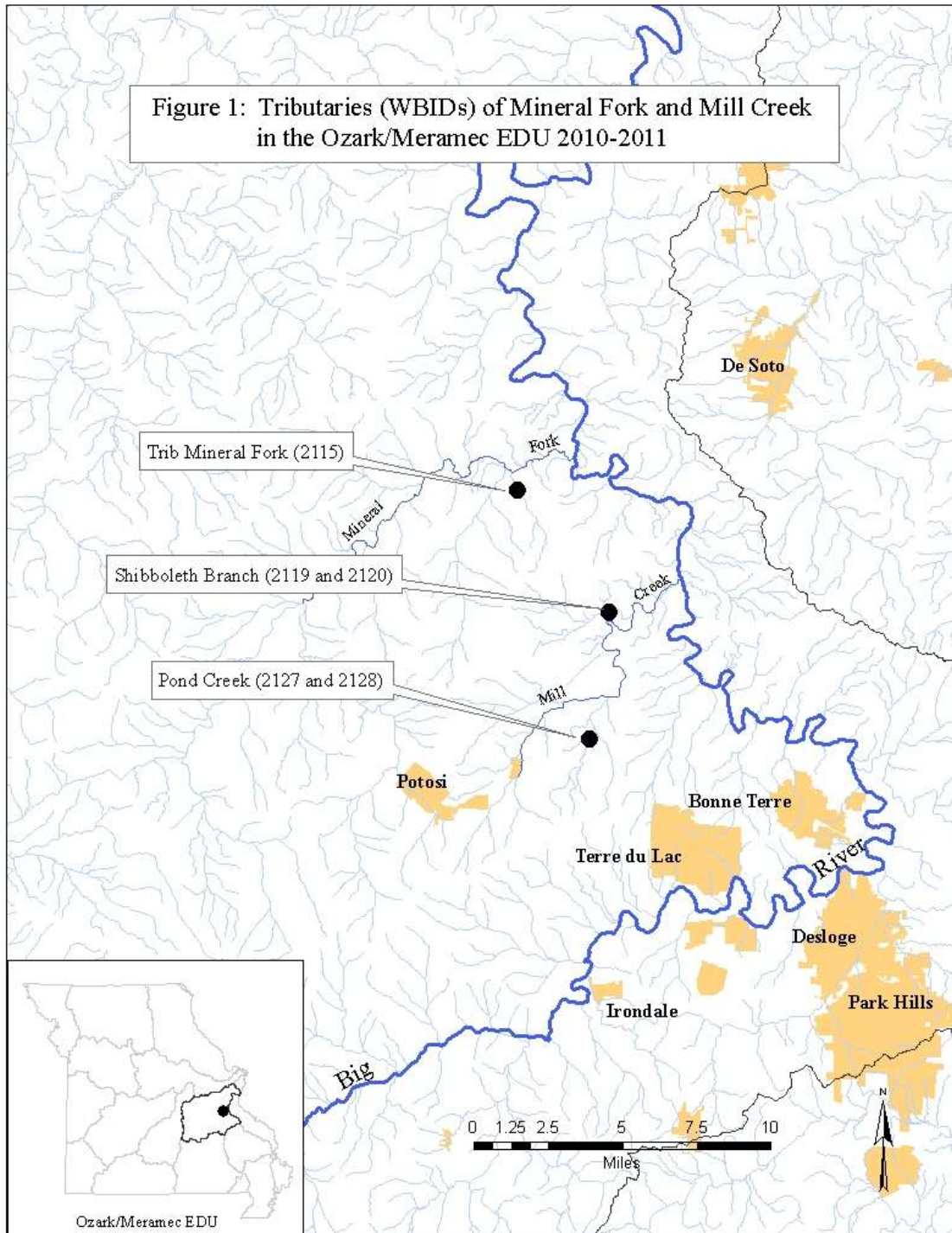
2.2.1 Ecological Drainage Unit

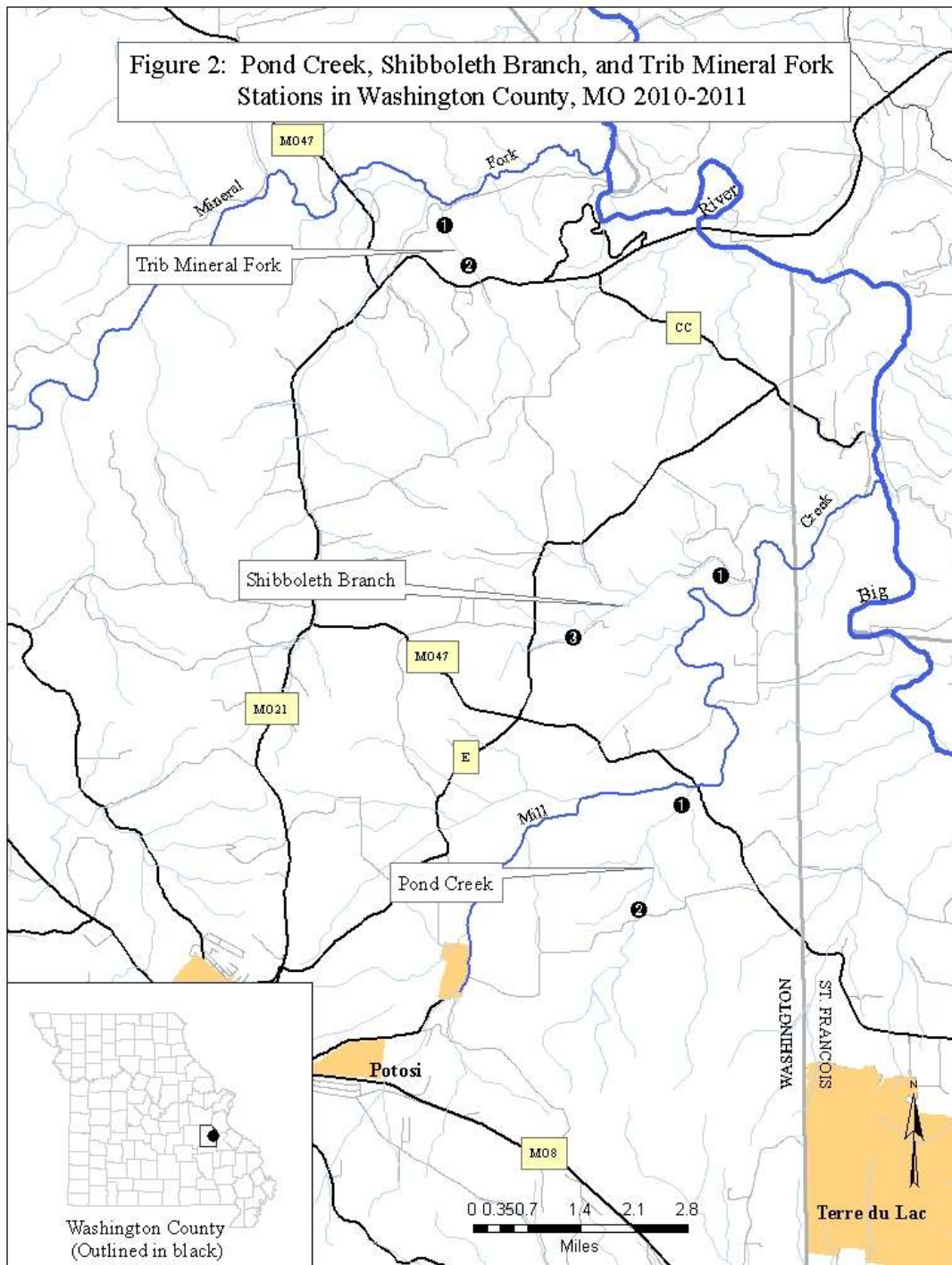
The tributaries and controls are located within the Ozark/Meramec EDU (Figure 1). Ecological Drainage Units are areas that are delineated and identified by their natural terrestrial physiographic division and major riverine watershed component. EDUs are further described in Sowa et al. (2007). Similar-size streams within an EDU are expected to contain similar habitat conditions and aquatic communities. Comparisons of habitat, biological and physicochemical results between test streams and references or similar-size control streams within the same EDU should then be appropriate.

Table 1
Locations and Descriptions of Tributaries 2010 and Control Streams 2009

Station	County	Location	Description; WBID	Purpose; Class
Pond Creek #2	Washington	NE ¼ sec.3, T. 37 N., R. 3 E. E703768 N4203267	Downstream Pond Creek Road; 2128	Test; C
Pond Creek #1	Washington	NW¼ sec. 35, T. 38 N., R. 3 E. E704861 N4205929	Upstream confluence with Mill Creek; 2127	Test; P
Shibboleth Branch #3	Washington	NE¼ sec. 21/NW sec. 22, T. 38 N., R. 3 E. E702030 N4209111	Apx 0.25 miles east of Hwy E, Powder Lake Spg. Rd; 2120	Test; C
Shibboleth Branch #1	Washington	NW¼ sec. 13, T. 38 N., R. 3 E. E705671 N4210490	Downstream bridge Johnson Road; 2119	Test; P
Trib. Mineral Fork #2	Washington	NW¼ sec. 28, T. 39 N., R. 3 E. E700345 N4216889	Downstream US 21; 2115	Test; C
Trib. Mineral Fork #1	Washington	NE¼ sec. 29, T. 39 N., R. 3 E. E699717 N4217961	Upstream Dugout Rd 0.25 mile; 2115	Test; C
Brazil Creek*	Washington	NE¼ sec. 28, T. 38 N., R. 1 W. E672696 N4206120	Downstream USFS Brazil Creek Camp	Control; P
Courtois Creek*	Iron	SW¼ sec. 28, T. 35 N., R. 1 W. E672115 N4175783	Downstream CR80A @ Goodwater, MO	Control; U
East Fork Huzzah Creek*	Dent	SW¼ sec. 20, T. 34 N., R. 2 W. E659956 N4164882	Downstream LWB 2 miles S Boss, MO	Control; C
West Fork Huzzah Creek*	Dent	SW¼ sec. 15, T. 34 N., R. 3 W. E653573 N4166719	Downstream MO Hwy 32 Howes Mill, MO	Control; C
Shoal Creek*	Crawford	NW¼ sec. 22, T. 36 N., R. 2 W. E663955 N4187505	USFS-Big Shoal Creek Road , NE Davisville	Control; P

* = Sampled in 2009 study (MDNR 2009a) and used here for comparison.





2.2.2 Land Use Description

Land use was compared among test stations, controls (candidate references), and the Ozark/Meramec EDU using a 14-digit Hydrological Unit scale (**HUC-14**; Table 2). Percent land cover data were derived from Thematic Mapper satellite data collected between 2000 and 2004 and interpreted by the Missouri Resource Assessment Partnership (**MoRAP**).

Land use or cover should be considered when examining stream habitat assessment or biological assessment results between stations or with the EDU. Land cover was relatively similar between the tributaries and the control stations, as well as with the general land cover of the Ozark/Meramec EDU. Overall, two land uses were dominant at the tributaries, controls, and the EDU. All tributaries, controls, and the EDU, in general had a high percentage of forest cover. The percentage of grassland cover was similar among most tributaries but was slightly less than the overall EDU. Therefore, general land use should not interfere with comparisons of results among stations or streams.

Table 2
Percent Land Use in the Tributaries, Control (Candidate Reference) Stations,
and the Ozark/Meramec EDU

Stations	HUC-14	Urban	Crops	Grass	Forest	Wetland	Open-water
Pond Creek #2, #1	071401040 80002	6	0	15	73	1	1
Shibboleth Branch #3, #2, #1	071401040 80002	6	0	15	73	1	1
Trib. Mineral Fork #2, #1	071401040 40003	1	0	10	83	2	1
Brazil Creek #1	071401020 50005	0	0	15	83	0	0
Courtois Creek #1	071401020 40001	1	0	8	86	0	0
East Fork Huzzah Creek #1	071401020 30001	0	0	17	80	0	0
West Fork Huzzah Creek #1	071401020 30001	0	0	17	80	0	0
Shoal Creek #1	071401020 30004	0	0	17	80	0	0
Ozark/Meramec EDU	--	4	1	27	62	0	0

2.3 Stream Habitat Assessment Project Procedure

The standardized Stream Habitat Assessment Project Procedure (SHAPP) was followed as described for riffle/pool prevalent streams (MDNR 2010c). According to the SHAPP, the quality of an aquatic community is based on the ability of the stream to support the aquatic community. If SHAPP scores at test stations are $\geq 75\%$ of the mean control scores, the stream habitat at the test station is considered to be comparable to the control streams. SHAPPs conducted at Brazil, Courtois, East Fork Huzzah, West Fork Huzzah, and Shoal creeks were used as controls (Table 1; MDNR 2009a). The SHAPP scores for Pond Creek, Shibboleth Branch, and the control streams are from the earlier study (MDNR 2009a). Trib. Mineral Fork was assessed during the fall 2010 of this study. Each stream habitat assessment score from the tributaries was compared as a percentage of the mean SHAPP control scores.

2.4 Biological Assessment

Sampling was conducted as described in the MDNR Semi-quantitative Macroinvertebrate Stream Bioassessment Project Procedure (SMSBPP), MDNR 2010b). Biological assessments consist of macroinvertebrate community and physicochemical water collection and analyses. Primary and secondary metrics were examined and are grouped by season, watershed, and station.

2.4.1 Macroinvertebrate Sampling and Analyses: Primary Metrics

Macroinvertebrates were sampled from multiple habitats as described in the SMSBPP (MDNR 2010b). The tributaries, references, and similar-size controls are considered riffle/pool dominant streams. As such, coarse substrate (**CS**; riffle), non-flowing water over depositional substrate (**NF**), and root mat (**RM**) habitats were sampled. Macroinvertebrates were subsampled in the WQMS lab according to the SMSBPP and identified to specific taxonomic levels in order to standardize calculation of the metrics (MDNR 2010b; MDNR 2010d).

Primarily, analyses of the macroinvertebrate community consisted of examination of Macroinvertebrate Stream Condition Index (**MSCI**) scores and the individual metrics that were used to generate the scores (MDNR 2010b).

An MSCI is a qualitative rank measurement of a stream's aquatic biological integrity (Rabeni et al. 1997). The MSCI was further refined for biological criteria reference streams (**BIOREF**) within each EDU in Biological Criteria for Perennial/Wadeable Streams (MDNR 2002), where comparisons are made between test streams and a BIOREF scoring range generated from data collected from wadeable/perennial reference streams. A station's MSCI score ultimately represents the ability of the stream to support the designated beneficial use for the protection of warm-water aquatic life (**AQL**).

An MSCI score is a compilation of rank scores that are assigned to individual biological metric scores as measures of biological integrity compared to BIOREFs. Four primary

biological metrics were compared to respective BIOREF scoring ranges and were used to calculate the MSCI per station: 1) Taxa Richness (**TR**); 2) Ephemeroptera/Plecoptera/Trichoptera Taxa (**EPTT**); 3) Biotic Index (**BI**); and 4) Shannon Diversity Index (**SDI**). Metric scores are compared to the BIOREF scoring range (BIOREF Scoring Table) and rank scores (5, 3, 1) are assigned to each metric. Rank scores were compiled and the MSCI was completed for each station. The MSCI scores are interpreted as follows: 20-16 = full support of AQL; 14-10 = partial support of AQL; and 8-4 = non-support of the AQL beneficial use designation. MSCI scores were compared among stations and grouped by season (Tables 4 and 5).

Individual biological metrics for each station were compared to the BIOREF scoring range to identify the level of integrity for each metric. Variations in the metrics may help identify how a community is affected and determine a potential source of impairment.

The MSCI scores of the tributaries were compared to scores based on criteria developed using control streams that were of similar size to the test streams. BIOREF streams are generally larger than the tributaries, and the macroinvertebrate communities may be different in smaller streams than larger ones. Therefore, a group of similar size control streams was chosen using methods similar to those used when selecting BIOREF streams. The earlier study of tributaries of Mill Creek and Mineral Fork (MDNR 2009a) suggested that the smaller control streams had fewer taxa, seasonally fewer EPTT, more sensitive taxa, and less diverse macroinvertebrate communities. A “Control Criteria” scoring range was generated for each season using the similar size control streams (Tables 4a and 5a). The control criteria were generated using the same methods that are outlined in the SMSBPP for the larger BIOREF streams. Comparisons were made between the BIOREF and control criteria MSCI scores (**ΔMSCI**), individual metric scores, and the biological support category (**ΔSupport**). A change in the MSCI score suggests that stream size was important in determining the support category, and describes the quality of the tributary compared to other streams of similar size.

2.4.2 Macroinvertebrate Analyses: Secondary Metrics

Secondary metrics are those that may highlight or support findings of the primary metrics. Two secondary metrics were examined and are explained here, which include percent sensitive taxa, and dominant macroinvertebrate families.

“Percent Sensitive Taxa” is a measure that shows the distribution of intolerant sensitive and tolerant taxa in the community composition based on BI values. The BI values range from 0 to 10 and describe the ability of an aquatic organism to tolerate organic pollution. Percentages of the total number of individuals in the subsample are calculated above and between the 90th, 75th, 50th, 25th and below the 25th percentile of the range, on a scale of one to ten. This breakdown provides a distribution of tolerant/intolerant taxa within the test stream communities, which allows for comparison with the BIOREF BI distribution within the EDU. This measure may help to explain sources of impairment by documenting shifts in the tolerance of the macroinvertebrate community. The second

biological analysis included is an examination of the “dominant macroinvertebrate families” (DMFs) per station. The seven most abundant DMFs for each station are listed as a percentage of the total number of individuals in the sample. Dominance by certain families may help identify the type and source of impairment. A more detailed taxa list is shown in the Macroinvertebrate Bench Sheet Report (Appendix A). The presence, absence, and abundance of certain species may help identify a type and source of impairment.

2.4.3 Physicochemical Water Sampling and Analyses

Physicochemical water samples were handled according to the applicable MDNR, ESP Standard Operating Procedures (SOP) and/or Project Procedures (PP) for sampling and analyzing physicochemical water samples. Results for physicochemical water variables were examined by season, watershed, and station. Stream bottom pore water samples were collected using peepers and analyzed by CAS.

Physicochemical water parameters consisted of field measurements and grab samples that were returned to the ESP environmental laboratory. Water was sampled according to the SOP MDNR-ESP-001 Required/Recommended Containers, Volumes, Preservatives, Holding Times, and Special Sampling Considerations (MDNR 2011). All samples that were transported to ESP were kept on ice. Temperature (°C), pH, conductivity (µS/cm), dissolved oxygen (mg/L), and discharge (cubic feet per second-cfs) were measured *in situ*. The ESP’s CAS in Jefferson City, Missouri conducted analyses for ammonia as nitrogen (NH₃-N; mg/L), nitrate+nitrite as nitrogen (NO₃+NO₂-N; mg/L), total nitrogen (TN; mg/L), chloride (Cl; mg/L), total phosphorus (TP; mg/L), and non-filterable residue (NFR; mg/L). Turbidity (nephelometric turbidity unit, NTU) was measured and recorded in the WQMS biology laboratory.

Test station physicochemical water parameters were compared to Missouri’s Water Quality Standards (WQS; MDNR 2010e). Interpretation of acceptable limits within the WQS may be dependent on a stream’s classification and its beneficial use designation (MDNR 2010e). Furthermore, acceptable limits for parameters may be dependent on the rate of exposure. These exposure or toxicity limits are based on the lethality of a toxicant given long-term (chronic toxicity) or short-term exposure (acute toxicity).

2.4.4 Discharge

Stream discharge was measured using a Marsh-McBirney Flowmate 2000™ flow meter at each station. Velocity and depth measurements were recorded at each station according to SOP MDNR-ESP-113 Flow Measurement in Open Channels (MDNR 2010a).

2.5 Dissolved Metals

Water samples analyzed for dissolved metals were collected using two methods in this project. Surface water was collected as a grab sample. Pore water was collected using peepers.

2.5.1 Surface Water

Surface water samples were collected for dissolved metals during the fall and spring sample seasons. Water samples for dissolved metals analysis were filtered through a 0.45µm filter in the field. Chemical analysis was conducted to determine the concentrations of the following dissolved metals: barium, cadmium, calcium, cobalt, copper, lead, magnesium, nickel, and zinc. Hardness as CaCO₃ values were calculated to identify chronic and acute metals toxicity concentrations as listed in Missouri's Water Quality Standards (MDNR 2010e).

2.5.2 Pore Water

Peepers (Serbst et al. 2003; Brumbaugh et al. 2002, 2007) were used *in situ* to collect samples for substrate pore water dissolved metals analysis. Materials used to construct the peepers were donated by the USGS's Columbia Environmental Research Center (CERC) in Columbia, Missouri. Peepers were prepared and deployed as described in Brumbaugh et al. (2007). Peepers were deployed at Trib. Mineral Fork #2, #1, and Shibboleth Branch #1 on September 21, 2010, and Shibboleth Branch #3, Pond Creek #2 and #1 on September 22, 2010. Peepers were buried in the substrate to a depth of approximately two inches in areas near the head of riffles as described by Brumbaugh et al. (2007). All samplers were retrieved October 14, 2010 except Pond Creek #2 samplers; both peepers appeared to have been removed from the deployment location. Water samples were analyzed for dissolved barium, cadmium, calcium, cobalt, copper, lead, nickel, and zinc. Hardness as CaCO₃ was calculated using calcium and magnesium concentrations according to APHA (1998). Results were then compared to Missouri Water Quality Standards (MDNR 2010e).

If heavy metals concentrations are elevated they may be developed into pore-water toxicity units (PWTU; USEPA 2005; Besser et al. 2009a, 2009b; MacDonald et al. 2009; Allert et al. 2011) and compared to threshold levels developed by MacDonald et al. (2009). A PWTU is the pore water dissolved metal concentration divided by the hardness dependent chronic level water quality standard. Chronic metals concentrations are listed in the Missouri Water Quality Standards (MDNR 2010e). A PWTU under 1.0 can be expected to be non-toxic (Besser et al. 2009b). The PWTUs may be summed (Σ PWTU; Besser et al. 2009a) to examine potential toxicity from metals mixtures and may be compared to pore water toxicity thresholds (MacDonald et al. 2009). The threshold value used is called the T10 threshold value, which corresponds to a 10% reduction in survival or biomass of the toxicity test organism. The Σ PWTU threshold value for divalent metals, which includes cadmium, lead, and zinc, is 1.03. Above this threshold the sample is expected to be toxic to benthic organisms.

A field blank was prepared to test sampling influences on the peepers. The field blank was taken to all sites in a sealed container in a cooler with ice for deployment and

retrieval. Prior to deployment the peepers were kept in ultra-pure water as described by Brumbaugh et al. (2007). During deployment, the container was placed in a cooler and kept in a refrigerator with a constant temperature near 3°C during deployment. The field blank and test peepers were capped in the field at the conclusion of the sample period. All samples were placed in separate plastic bags, placed on ice, and transported to ESP. Dissolved metals were analyzed by MDNR's CAS using applicable SOPs.

2.6 Fine Sediment

Instream deposits of fine sediment (i.e. particle size ca. <2 mm) were estimated for percent coverage per area and characterized for composition of total recoverable metals (TR; µg/kg). The CAS of ESP conducted metals character analyses.

2.6.1 Fine Sediment Percent Coverage Estimation

The relative percentage of fine sediment coverage was visually estimated for each station. The visual estimates were conducted within a 0.25 m² metal quadrat that was randomly located in sample areas called grids (Figure 3). Each station contained three grids. This method allowed for estimation and comparison of the relative coverage of benthic fine sediment among stations.

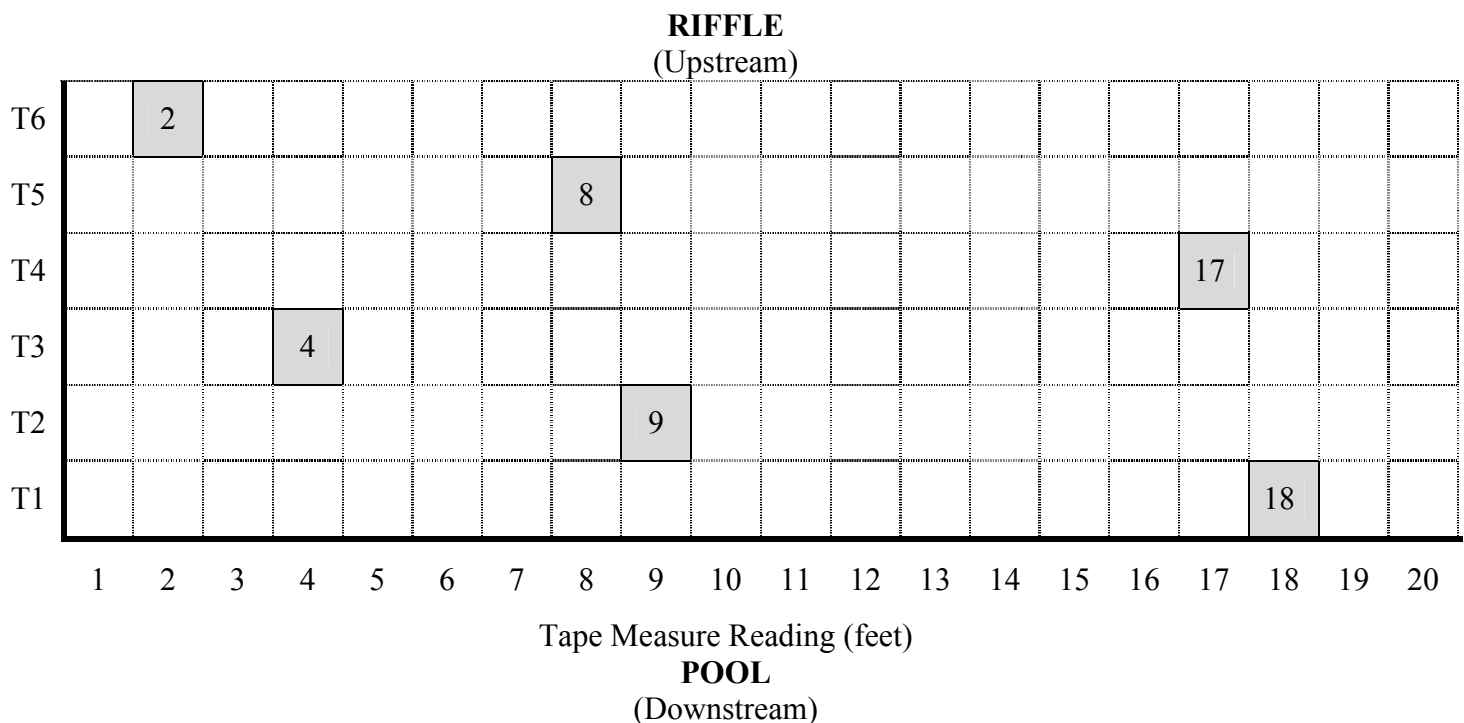
To ensure sampling method uniformity, grids were located at the downstream margins of riffles or runs and the upstream margin of pool habitats in areas of relatively laminar flow. Grid placement was similar to previous fine sediment assessment projects conducted by the MDNR WQMS, including Flat River (MDNR 2001) and Upper Big River (MDNR 2003). Water velocity was ≤0.5 feet per second, which allows fine sediment-sized particles to settle from transport after high flow events, according to the Hjølstrom Diagram for threshold transport and settling velocities (Hjølstrom 1939). A Marsh-McBirney flow meter was used to determine maximum velocity within the proposed grid. Depths did not exceed three feet. Grids excluded eddies, bends, and areas downstream of vegetation or large obstructions that may cause turbulent flow.

Once a suitable area was selected, a virtual *grid* was constructed (Figure 4). A 100 foot tape measure was anchored and stretched across the stream. The tape served as the downstream border of a virtual grid of six contiguous *transects*. Each transect was 12 inches wide (equal to the grid width) and its location was determined by measuring upstream from the 100 foot tape with a retractable tape measure. A random number, equating to a one foot increment, was drawn to determine where the *quadrat* was placed in the first transect. The quadrat was placed on the substrate with the downstream edge contacting the downstream edge of the first transect. Two observers estimated and recorded the percent of fine sediment within the quadrat. The estimates were accepted and recorded if the two observations were within a ten percent margin of error. If estimates differed by more than ten percent, they were rejected and observations were repeated until the estimates were within the acceptable margin of error. A second random number was then drawn and the quadrat was placed in the second transect upstream (twelve inches farther from the hundred foot tape) for the next observation.

This process continued until fine sediment was estimated at random locations within each of the six quadrats (one per transect).

A mean of the two estimates was calculated for each transect and was used for later analyses (Table 15). The coverage data were examined using Analysis of Variance on Ranks, with multiple comparison procedures if significant differences ($p < 0.05$) were detected between tributaries and controls (SigmaStat version 3.5 2006).

Figure 3: Virtual grid of transects (T) and quadrats (boxes in gray, numbered) used to estimate percent fine sediment. Example: stream twenty feet wide; quadrat placement was based on random numbers (e.g. 18, 9, 4, 17, 8, 2)



2.6.2 Fine Sediment Character and Analyses

Fine sediment was sampled for total recoverable metals within each station. One 2-ounce jar of representative fine sediment was collected from the substrate in each grid (see Section 2.5.1). Three samples per station were composited into one 8-ounce glass jar per station. The fine sediment was subsampled and analyzed by CAS for total recoverable cadmium, lead, and zinc. Individual concentrations and mixture of metals thresholds were compared to thresholds levels (mg/kg).

Individual metals concentrations were compared to PEC (MacDonald et al. 2000). A PEC is the level of a contaminant above which harmful effects are likely to be observed. MacDonald et al. (2000) found PECs to be reliable for ten metals (including cadmium, lead, and zinc) for classifying sediments as nontoxic or toxic. PEC for lead is 128 mg/kg

dry weight, the PEC for cadmium is 4.98 mg/kg, and the PEC for zinc is 459 mg/kg (MacDonald et al. 2000).

Individual metals were also examined using a probable effects concentration quotient (**PEQ**; MacDonald et al. 2000, 2009; Ingersoll et al. 2001, 2002, 2009; Besser et al. 2008, 2009a). The PEQ is the total recoverable concentration divided by that metal's respective PEC (MacDonald et al. 2000). A PEQ greater than 1.0 may be associated with an increased risk of toxicity (Besser et al. 2009a).

The effects from a mixture or combination of metals may be accounted for using a sum of PEQs (Σ PEQ) or developing the mean PEQ. The Σ PEQ (Besser et al. 2009a; MacDonald et al. 2009; Allert et al. 2011) is the sum of PEQs for all three metals that accounts for potential effects from cadmium, lead, and zinc mixtures. The mean PEQ is the Σ PEQs divided by the number of metals in the mixture (Long et al. 1998; MacDonald et al. 2000; Ingersoll et al. 1998, 2001, 2002, 2009; Besser et al. 2008, 2009b).

The Σ PEQ and mean PEQ may then be compared to threshold levels. Threshold levels were developed by MacDonald et al. (2009) for both the Σ PEQ and mean PEQ that estimate the risk of metals mixtures on benthic invertebrates. Although the sum and mean methods each are effective and accurate, both are included here. The thresholds for cadmium, lead, and zinc are Σ PEQ=7.92 and mean PEQ=1.11. Metals toxicity above these thresholds is considered high risk to the population.

2.7 Quality Control

Quality control was conducted in accordance with applicable MDNR SOPs. Macroinvertebrate community and water physicochemical variables were duplicated for every 10 stations sampled.

3.0 Results

Results are grouped by 1) stream habitat assessment; 2) biological assessment, which includes macroinvertebrate community and water quality; 3) dissolved metals sections, which includes surface and pore water; and 4) fine sediment coverage estimations and characterization. Trends and exceptional results are highlighted.

3.1 Stream Habitat Assessment

Stream habitat assessment scores were compared as a percentage of the mean of SHAPP control scores (Table 3). All stations exceeded the 75 percent similarity threshold with the mean of SHAPP controls. Mill Creek and control SHAPPs were conducted in the earlier study (MDNR 2009a) and their results are repeated here for discussion. The range of test stream SHAPP scores was from 81 percent to greater than 100 percent of the control scores. Trib. Mineral Fork SHAPPs were conducted during this study. The habitat scores were similar between stations and the percentages were similar to the mean of controls.

3.2 Biological Assessment

The biological assessment, which includes macroinvertebrate community analyses and physicochemical water quality analyses, are found in this section. Secondary metrics and pore water results are also included in this section. Results are grouped by season, watershed, and station where applicable.

Table 3
Stream Habitat Assessment Project Procedure (SHAPP) Scores, and
Comparisons with SHAPP Control Streams

Station	SHAPP Score	Percent Mean of Controls
Pond Creek #2 *	139	89
Pond Creek #1 *	162	>100
Shibboleth Branch #3*	134	86
Shibboleth Branch #1*	126	81
Trib. Mineral Fork #2	130	83
Trib. Mineral Fork #1	138	88
Brazil Creek #1* (control)	161	
Courtois Creek #1* (control)	146	
West Fork Huzzah Creek #1 * (control)	169	
East Fork Huzzah Creek #1* (control)	152	
Shoal Creek #1* (control)	151	
Mean of Controls	156	

* Results from 2008 study (MDNR 2009a).

3.2.1 Macroinvertebrate Community Analyses

Macroinvertebrate community analyses include examination of MSCI scores and individual metrics compared to BIOREF criteria. Since these tributaries are smaller, and the scores may potentially be influenced by stream size, MSCI scores and individual metrics are also compared to similar size control criteria. Secondary metrics, such as percent sensitive taxa and dominant macroinvertebrate families are also included in this section. Each section is grouped by season, watershed, and station.

3.2.1.1 MSCI – Fall 2010

One stream in the Mill Creek watershed (Shibboleth Branch #3) was partially supporting in the fall of 2010 (Table 4). The Shibboleth Branch #3 partially supporting MSCI score was due to lower TR, EPTT and SDI metrics. Shibboleth Branch #1 was fully supporting, but had a high biological integrity (**BI**). Pond Creek #2 had an elevated BI and lower SDI, but maintained full support. Downstream, Pond Creek #1 scored the optimum 20 and was fully supporting. Both stations in the Mineral Fork watershed were fully supporting in the fall of 2010 (Table 4). Trib. Mineral Fork #2 had an MSCI score of 18, which placed

it in the full support category. The score was less than optimum due to an elevated BI. Trib. Mineral Fork #1 was fully supporting of its beneficial use designation, with an optimum MSCI score of 20.

Table 4
Biological Criteria (BIOREF) Metric Scores, Biological Support Category, and Macroinvertebrate Stream Condition Index (MSCI) Scores for Tributaries, Fall 2010

Stream and Station Number	Sample No.	TR	EPTT	BI	SDI	MSCI	Support
Pond Creek #2	1004018	94	25	6.6	2.84	16	F
Pond Creek #1	1004019	90	24	5.7	3.29	20	F
Shibboleth Branch #3	1004020	72	13	5.5	2.79	14	P
Shibboleth Branch #1	1004017	91	23	6.1	3.44	18	F
Trib. Mineral Fork #2	1004015	100	23	6.2	3.43	18	F
Trib. Mineral Fork #1	1004016	92	23	5.1	3.49	20	F
BIOREF Score=5	--	>79	>21	<5.8	>3.09	20-16	Full
BIOREF Score=3	--	79-39	21-11	5.8-7.9	3.09-1.55	14-10	Partial
BIOREF Score=1	--	<39	<11	>7.9	<1.55	8-4	Non

MSCI Scoring Table (bottom) developed from BIOREF samples (n=7); TR=Taxa Richness; EPTT=Ephemeroptera, Plecoptera, Trichoptera Taxa; BI=Biotic Index; SDI=Shannon Diversity Index; **Bold**=less than optimum BIOREF score.

Because test streams in this study were smaller than BIOREF streams, criteria were developed using the group of similar-sized control streams to compare with the test stations (Table 4a and 5a). These control criteria were another measure to determine if the size of these streams affected the MSCI scores.

The control criteria were compared to the fall metric scores of tributaries in the Mill Creek watershed (Table 4a). Although the Pond Creek #2 MSCI score did not change, Pond Creek #1 decreased from 20 to 18 due to a higher BI value. The Shibboleth Branch #3 score decreased slightly, again due to a higher BI. Shibboleth Branch #1 metric scores were not different between the BIOREF and control criteria.

The two Trib. Mineral Fork stations responded in different ways to control stream criteria (Table 4a). Trib. Mineral Fork #2 had no changes in the MSCI or individual metric scores. The higher BI value at Trib. Mineral Fork #1 resulted in a lower MSCI score.

Regardless, both Trib. Mineral Fork stations maintained full support of the AQL beneficial use in the fall.

Table 4a
Fall 2010 Control Criteria Metric Scores, Biological Support Category, and Macroinvertebrate Stream Condition Index (Δ MSCI) Scores, Highlighting Changes in Scores Using Similar Size Control Criteria from Fall 2008

Stream and Station Number	Sample No.	TR	EPTT	BI	SDI	Δ MSCI	Δ Support
Pond Creek #2	1004018	94	25	6.6	2.84	16 (NC)	F (NC)
Pond Creek #1	1004019	90	24	5.7	3.29	20→18	F (NC)
Shibboleth Branch #3	1004020	72	13	5.5	2.79	14→12	P (NC)
Shibboleth Branch #1	1004017	91	23	6.1	3.44	18 (NC)	F (NC)
Trib. Mineral Fork #2	1004015	100	23	6.2	3.43	18 (NC)	F (NC)
Trib. Mineral Fork #1	1004016	92	23	5.1	3.49	20→18	F (NC)
Control Criteria Score=5	--	>75	>21	<5.1	>2.97	20-16	Full
Control Criteria Score=3	--	75-37	21-11	5.1-7.5	2.97-1.49	14-10	Partial
Control Criteria Score=1	--	<37	<11	>7.9	<1.49	8-4	Non

Control Criteria MSCI Scoring Table (in light gray) developed from control streams (n=5) in MDNR 2009a); TR=Taxa Richness; EPTT=Ephemeroptera, Plecoptera, Trichoptera Taxa; BI=Biotic Index; SDI=Shannon Diversity Index; **Bold**=less than optimum control criteria score; **Highlight**=change in value from original metric score; NC=No Change .

3.2.1.2 MSCI – Spring 2011

Several of the streams sampled in the Mill Creek watershed garnered partial support of the beneficial use for the protection of AQL in the spring of 2011 (Table 5). Pond Creek #2 was partially supporting with a score of 14, as a result of less than optimum EPTT, BI, and SDI scores. Pond Creek #1 was fully supporting despite a suboptimal EPTT score. Shibboleth Branch #3 was partially supporting, which resulted from lower TR, EPTT, and SDI metric scores. Despite having a high TR value, Shibboleth Branch #1 also had a partially supporting MSCI score due to suboptimal EPT, SDI, BI scores.

The Trib. Mineral Fork #1 MSCI score was partially supporting in the spring due to suboptimal TR, EPTT, and SDI scores (Table 5). Trib. Mineral Fork #2 was fully supporting, but had a suboptimal EPTT score and a BI value higher than the optimum

range. Trib. Mineral Fork #1 was partially supporting with lower TR and SDI values than upstream, yet BI was in the optimum range.

Table 5
Biological Criteria (BIOREF) Metric Scores, Biological Support Category, and Macroinvertebrate Stream Condition Index (MSCI) Scores for Tributaries, Spring 2011

Stream and Station Number	Sample No.	TR	EPTT	BI	SDI	MSCI	Support
Pond Creek #2	110323	93	20	6.2	2.84	14	P
Pond Creek #1	110324	93	27	5.7	3.37	18	F
Shibboleth Branch #3	110328	75	19	5.1	3.05	14	P
Shibboleth Branch #1	110325	110	29	6.2	3.29	14	P
Trib. Mineral Fork #2	110327	95	24	5.9	3.35	16	F
Trib. Mineral Fork #1	110326	89	27	5.7	3.11	14	P
BIOREF Score=5	--	>92	>29	<5.8	>3.33	20-16	Full
BIOREF Score=3	--	92-46	29-15	5.8-7.9	3.33-1.67	14-10	Partial
BIOREF Score=1	--	<46	<15	>7.9	<1.67	8-4	Non

MSCI Scoring Table (bottom) developed from BIOREF samples (n=6); TR=Taxa Richness; EPTT=Ephemeroptera, Plecoptera, Trichoptera Taxa; BI=Biotic Index; SDI=Shannon Diversity Index
Bold=less than optimum BIOREF score.

Control stream criteria again were used to calculate spring MSCI scores for tributaries in the Mill Creek watershed (Table 5a). The Pond Creek #2 biological metric scores and overall MSCI score were the same for both sets of criteria. Using control stream criteria, the Pond Creek #1 EPTT biological metric had the highest possible score; however, the BI score decreased to partially supporting, which resulted in no change in the MSCI. Similarly, the Shibboleth Branch #3 MSCI had no change, despite changes in the BI and SDI biological metric scores. Using BIOREF criteria the Shibboleth Branch #1 MSCI score was partially supporting; however, with control stream criteria both the EPTT and SDI biological metrics were in the fully supporting range, which changed the overall MSCI score from 14 to 18 (Table 5a).

Scores for each of the biological metrics and the MSCI for Trib. Mineral Fork #2 were the same for BIOREF and control stream criteria in the spring (Table 5a). Each of the Trib. Mineral Fork #1 biological metric scores, however, changed when using the control

stream criteria and resulted in the MSCI score increasing from 14 to 18. All metrics were affected as the TR, EPTT, and SDI increased to the optimum score, whereas the BI score decreased below the optimum range.

Both Trib. Mineral Fork stations #2 and #1 were fully supporting; however, station #2 had a suboptimum BI in the fall. The distribution at station #2 showed approximately 41 percent of the macroinvertebrate population with a BI above 7.5. By contrast, station #1 had only 20 percent in the same range. The BI distribution above 7.5 in the EDU was also approximately 20 percent.

Table 5a
Spring 2011 Control Criteria Metric Scores, Biological Support Category, and Macroinvertebrate Stream Condition Index (ΔMSCI) Scores, Highlighting Changes in Scores Using Similar Size Control Criteria from Spring 2009

Stream and Station Number	Sample No.	TR	EPTT	BI	SDI	ΔMSCI	Δ Support
Pond Creek #2	110323	93	20	6.2	2.84	14 (NC)	P (NC)
Pond Creek #1	110324	93	27	5.7	3.37	18 (NC)	F (NC)
Shibboleth Branch #3	110328	75	19	5.1	3.05	14 (NC)	P (NC)
Shibboleth Branch #1	110325	110	29	6.2	3.29	14→18	P→F
Trib. Mineral Fork #2	110327	95	24	5.9	3.35	16 (NC)	F (NC)
Trib. Mineral Fork #1	110326	89	27	5.7	3.11	14→18	P→F
Control Criteria Score=5	--	>81	>26	<4.5	>3.00	20-16	Full
Control Criteria Score=3	--	81-41	26-13	4.5-7.3	3.00-1.50	14-10	Partial
Control Criteria Score=1	--	<41	<13	>7.3	<1.50	8-4	Non

MSCI Scoring Table (in light gray) developed from control streams (n=5); TR=Taxa Richness; EPTT=Ephemeroptera, Plecoptera, Trichoptera Taxa; BI=Biotic Index; SDI=Shannon Diversity Index; **Bold**=less than optimum control criteria (MDNR 2009a) score; **Highlight** = change in value from original metric score

3.2.1.3 Percent Sensitive Taxa – Fall 2010

The percent sensitive taxa metric was calculated for tributaries in the fall and compared among stations as well as with the EDU (Table 6). Pond Creek stations #2 and #1 were both fully supporting of the AQL designation in the fall. Pond Creek #2 had an elevated BI, which was the result of over 51 percent of taxa in the sample with a BI value above 7.5. Pond Creek #1 had only 21 percent of the population above the BI of 7.5.

Shibboleth Branch #3 was partially supporting, with an optimum BI score and approximately 62 percent of taxa with a BI greater than 5. Shibboleth Branch #1 was fully supporting, yet it had a suboptimum BI score resulting from 73 percent of taxa with a BI above 5. By comparison, the EDU as a whole had approximately 70 percent of taxa above 5 as well.

The Percent Sensitive Taxa distribution for fall 2010 is illustrated in Table 6. Trib. Mineral Fork station #2 showed approximately nearly 80 percent of the population had a BI value above 5 and subsequently had a less than optimum BI value. Trib. Mineral Fork #1 had approximately 58 percent above 5 and had an optimum BI. The population in the EDU by contrast was composed of approximately 70 percent above 5.

Table 6
Percent Sensitive Taxa (based on BI scoring range) by Station and EDU, Fall 2010

Stream/Station	Sample Numbers	<2.5	2.5 to <5	5 to <7.5	7.5 to <9	>9
Pond Creek 2	1004018	2.73	10.94	34.52	50.10	1.71
Pond Creek 1	1004019	2.56	29.92	42.48	21.16	3.88
Shibboleth Branch 3	1004020	1.51	34.55	49.76	9.98	4.20
Shibboleth Branch 1	1004017	5.87	14.88	44.97	30.89	3.38
Trib. Mineral Fk 2	1004015	4.45	16.37	37.47	37.33	4.38
Trib. Mineral Fk 1	1004016	9.77	31.82	38.06	18.27	2.08
EDU	--	10.56	18.75	47.81	20.14	2.75

Bold=partial support; **Highlight**=high BI

3.2.1.4 Percent Sensitive Taxa – Spring 2011

The percent sensitive taxa were calculated for spring 2011 results (Table 7). Mill Creek watershed stations were compared within each stream and with the EDU. Pond Creek #2, which had a partially supporting MSCI score, had a suboptimal BI score. Alternatively, Pond Creek #1 had a fully supporting MSCI score and a BI score in the optimal range. Over 48 percent of the Pond Creek Station #2 sample was made up of taxa with BI values over 7.5, as opposed to Pond Creek #1 which had approximately 25 percent over 7.5. Shibboleth Branch #3 had a partially supporting MSCI score with an optimal BI score. Shibboleth Branch #1 also had a partially supporting MSCI score, but with a suboptimal BI score. Shibboleth Branch #3 had only six percent above 7.5, whereas the community at #1 was made up of over 30 percent above 7.5. The EDU had about 18 percent above 7.5.

The distribution of taxa sensitivity for spring samples is presented for Trib. Mineral Fork stations in Table 7. Trib. Mineral Fork #2 had a fully supporting MSCI score, with a suboptimal BI score. The downstream Trib. Mineral Fork #1 station had a partially supporting MSCI score with an optimal BI score. Approximately 80 percent of the station #2 population had a BI above 5 and station #1 had approximately 70 percent with a BI above 5. The EDU contained approximately 70 percent above 5.

Table 7
Percent Sensitive Taxa (based on BI scoring range) by Station and EDU, Spring 2011

Stream/Station	Sample Numbers	<2.5	2.5 to <5	5 to <7.5	7.5 to <9	>9
Pond Creek 2	1004018	7.45	16.11	27.89	45.52	3.04
Pond Creek 1	1004019	8.74	12.85	52.73	24.87	0.82
Shibboleth Branch 3	1004020	2.38	45.42	46.09	5.29	0.82
Shibboleth Branch 1	1004017	8.16	7.28	53.28	29.68	1.60
Trib. Mineral Fk 2	1004015	3.28	14.58	55.06	24.56	2.51
Trib. Mineral Fk 1	1004016	8.22	21.16	35.74	34.11	0.78
EDU	--	12.08	13.02	56.72	16.34	1.84

Bold=partial support; **Highlight**=high BI

3.2.1.5 Dominant Macroinvertebrate Families – Fall 2010

The dominant macroinvertebrate family metric was used to describe the macroinvertebrate community composition for each tributary in the fall (Table 8). Pond Creek #2 was dominated by Caenidae and Chironomidae. Pond Creek #1 was also dominated by Caenidae, although with a lower relative percentage; Ephemerellidae also was among the dominant families at this station. Shibboleth Branch #3 had a partially supporting MSCI score and was dominated by Chironomidae (47.3 percent). Shibboleth Branch #1 was fully supporting and was dominated by Chironomidae (32.9 percent) and Caenidae (22.9 percent).

Trib. Mineral Fork #2 was dominated by Caenidae in the fall (Table 8). Trib. Mineral Fork #1 had fewer Caenidae than #2 and Heptageniidae was more abundant. Both Mineral Fork stations were fully supporting.

3.2.1.6 Dominant Macroinvertebrate Families – Spring 2011

The dominant macroinvertebrate families metric was used to describe the macroinvertebrate community composition for each tributary station in the spring (Table 9). Pond Creek #2, which had a partially supporting MSCI score, was dominated by Caenidae. Pond Creek #1 was dominated by Chironomidae and Caenidae. Shibboleth Branch #3 and #1 were both partially supporting and the samples were dominated by Chironomidae.

Trib. Mineral Fork #2 was dominated by about 44 percent Chironomidae in the spring (Table 9). Trib. Mineral Fork #1 was partially supporting with 28 percent Caenidae and 22 percent Chironomidae

Table 8
Dominant Macroinvertebrate Families (DMF) as a Percentage of the Total
Number of Individuals per Station for Tributaries, Fall 2010

Family	Pond Creek #2	Pond Creek #1	Shibboleth Branch #3	Shibboleth Branch #1	Trib. Mineral Fork #2	Trib. Mineral Fork #1
Caenidae	43.3	16.8	-	22.9	28.0	10.6
Chironomidae	19.3	13.0	47.3	32.9	22.7	26.8
Heptageniidae	4.4	9.3	23.8	6.1	4.5	17.5
Elmidae	4.0	12.4	3.4	11.2	13.6	14.5
Hydropsychidae	3.4	-	2.8	-	-	-
Empididae	2.9	-	-	-	-	-
Coenagrionidae	2.1	-	-	2.5	-	-
Ephemerellidae	-	14.4	-	-	-	-
Gomphidae	-	8.8	-	-	-	-
*Arachnoidea	-	3.2	-	-	-	-
Isonychiidae	-	-	-	3.0	-	-
Asellidae	-	-	-	2.8	3.5	-
Leptoceridae	-	-	4.1	-	-	-
Baetidae	-	-	2.3	-	-	-
Calopterygidae	-	-	1.9	-	-	-
Philopotamidae	-	-	-	-	3.7	4.7
Tubificidae	-	-	-	-	3.3	-
Psephenidae	-	-	-	-	-	2.3
Perlidae	-	-	-	-	-	2.3

Bold=Partial support; *= Order

3.2.2 Physicochemical Water Quality Analyses

General physicochemical water quality analyses are included in this section. General water quality includes results from grab samples and field measurements taken during each biological assessment visit. Interesting results or trends are highlighted. All results were within WQSs for both seasons (MDNR 2010e). Results are grouped by season, watershed, and station.

3.2.2.1 General Water Quality – Fall 2010

Overall water quality parameters for the Mill Creek tributaries were not remarkable in the fall, but a few trends were highlighted (Table 10). Chloride was present in detectable concentrations at all test stations, but it tended to be higher at the Trib. Mineral Fork sites. Total nitrogen and nitrate+nitrite-N were lowest at the Pond Creek stations. The highest total nitrogen concentrations were observed at the Trib. Mineral Fork stations.

Nitrate+nitrite-N at Shibboleth Branch #1 (0.12 mg/L) was more than twice the concentration observed at the upstream station (0.05 mg/L). Ammonia-N and total phosphorus were present at or near non-detectable concentrations at all stations. Among non-nutrient water quality parameters, conductivity and, as mentioned previously chloride were highest at the two Trib. Mineral Fork stations. Non-filterable Residue (**NFR**) also was higher at the Trib. Mineral Fork stations, with the Station #2 concentration (10.0 mg/L) being roughly twice as high as any of the remaining stations. Discharge, however, also was higher at this station. All water quality results listed in Table 10 were within WQSs (MDNR 2010e).

Table 9
Dominant Macroinvertebrate Families (DMF) as a Percentage of the Total
Number of Individuals per Station for Tributaries, Spring 2011

Family	Pond Creek #2	Pond Creek #1	Shibboleth Branch #3	Shibboleth Branch #1	Trib. Mineral Fork #2	Trib. Mineral Fork #1
Caenidae	41.8	22.4	-	20.7	20.4	30.8
Chironomidae	18.6	29.6	41.9	46.0	43.9	28.7
Simuliidae	6.0	-	16.9	-	2.7	6.9
Heptageniidae	5.9	6.7	17.3	4.9	2.2	5.9
Empididae	5.5	-	2.8	1.7	-	2.2
Tubificidae	2.5	-	-	-	-	-
*Arachnoidea	2.2	4.9	2.6	3.9	2.0	-
Ephemerellidae	-	9.9	-	-	-	-
Elmidae	-	4.5	-	6.4	7.9	9.6
Leuctridae	-	3.8	-	-	-	-
Hydropsychidae	-	-	2.1	-	-	-
Nemouridae	-	-	1.6	-	-	1.9
Isonychiidae	-	-	-	4.1	-	-
Pleuroceridae	-	-	-	-	3.2	-

Bold=Partial support; *=Order

3.2.2.2 General Water Quality – Spring 2011

Overall water quality parameters for the Mill Creek tributaries were not remarkable in the spring, but a few trends were highlighted (Table 11). Total nitrogen was present in similar concentrations at all test stations, with the exception that Pond Creek #1 and Shibboleth Branch #3 had somewhat lower levels. The Pond Creek #1 total nitrogen concentration (0.12 mg/L) was less than half of the upstream Pond Creek #2 sample (0.27 mg/L), which had the highest total nitrogen levels among all test stations. Nitrate+nitrite-N also were similar among stations, but was slightly higher (0.09 mg/L) at Shibboleth Branch #1 compared to the other sites. Ammonia-N and total phosphorus each were present in concentrations below detectable levels. Among non-nutrient water quality parameters, conductivity, and chloride were highest at the two Trib. Mineral Fork stations. Turbidity and NFR were similar among stations.

Table 10
Physicochemical Water Parameters for the Tributaries,
Fall 2010

Station Variable/Date	Pond Creek #2	Pond Creek #1	Shibboleth Branch #3	Shibboleth Branch #1	Trib. Mineral Fork #2	Trib. Mineral Fork #1
Sample Number	1006855	1006856	1006857	1006854	1006852	1006853
pH (Units)	8.2	8.2	8.3	8.5	8.3	8.2
Temperature (°C)	20.0	20.0	21.0	22.0	19.0	20.0
Conductivity (µS/cm)	318	430	321	442	531	517
Dissolved O ₂	8.04	8.23	7.27	7.87	7.99	6.81
Discharge (cfs)	0.49	0.89	0.61	3.80	6.56	0.56
NFR	5.0	5.0	<5.0	<5.0	10.0	6.0
Turbidity (NTUs)	2.08	0.73	1.52	3.10	7.14	0.95
Total Nitrogen	0.06	0.07	0.17	0.20	0.28	0.22
Nitrate+Nitrite-N	<0.01	0.01	0.05	0.12	0.07	0.05
Ammonia-N	<0.03	<0.03	<0.03	<0.03	<0.03	<0.03
Chloride	3.26	4.80	4.83	4.07	8.12	7.72
Total Phosphorus	0.01	<0.01	0.01	0.01	0.02	0.02

Units mg/L unless otherwise noted; **Bold**=notable or outside acceptable WQS range;
Highlight = High BI; **Bold station name**=partial support

Table 11
Physicochemical Water Parameters for the Tributaries,
Spring 2011

Station Variable/Date	Pond Creek #2	Pond Creek #1	Shibboleth Branch #3	Shibboleth Branch #1	Trib. Mineral Fork #2	Trib. Mineral Fork #1
Sample Number	1104220	1104176	1104180	1104177	1104179	1104178
pH (Units)	7.6	8.5	8.7	8.5	8.6	8.6
Temperature (°C)	13.0	13.0	9.0	17.0	9.0	9.0
Conductivity (µS/cm)	193	257	277	339	421	413
Dissolved O ₂	8.88	8.99	11.21	9.32	11.67	11.05
Discharge (cfs)	3.06	7.51	2.35	12.56	2.63	2.77
NFR	<5.0	<5.0	<5.0	<5.0	<5.0	<5.0
Turbidity (NTUs)	5.65	2.77	2.38	2.90	4.52	3.40
Total Nitrogen	0.27	0.12	0.16	0.20	0.22	0.20
Nitrate+Nitrite-N	0.05	0.03	0.04	0.09	0.05	0.02
Ammonia-N	<0.03	<0.03	<0.03	<0.03	<0.03	<0.03
Chloride	2.94	2.76	5.55	4.46	6.94	6.48

Total Phosphorus	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
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Units mg/L unless otherwise noted; **Bold**=noteable or outside acceptable WQS range;
Highlight = High BI; **Bold station name**=partial support

3.3 Dissolved Metals: Surface Water and Pore Water

All Mill Creek tributaries and Trib. Mineral Fork contained elevated concentrations of dissolved metals (i.e. barium, cadmium, lead, zinc) in the surface water during the fall and spring. Metals were considered elevated if the concentration was higher than the controls. Dissolved metals concentrations were below water quality standards (MDNR 2010e) for all surface water samples. Pore water samples for one test station, however, had dissolved metals concentrations that exceeded the standards (MDNR 2010e).

3.3.1 Surface Water – Fall 2010 and Spring 2011

Several Mill Creek tributaries fall surface water samples had dissolved metals that were detected in concentrations above the controls (Table 12). Pond Creek #2 and #1 samples contained barium and nickel in elevated or detectable concentrations; Shibboleth Branch #3 contained barium, lead, and nickel; and Shibboleth Branch #1 contained barium and nickel. Although these test stations had metals in concentrations above the control streams, none of the dissolved metals exceeded WQSs (MDNR 2010e) in the fall.

Dissolved metals were detected in surface water samples at Trib. Mineral Fork in the fall (Table 12). Trib. Mineral Fork #2 had elevated barium, nickel, and zinc. Trib. Mineral Fork #1 had barium and nickel in concentrations similar to the upstream station; however, the zinc concentration was similar to controls. None of the dissolved metals exceeded WQSs (MDNR 2010e) in the fall.

Dissolved metals were detected in the surface water of Mill Creek tributaries in concentrations above the controls in spring 2011 (Table 13). Pond Creek #2 contained dissolved barium and nickel concentrations above those found in the control streams. Pond Creek #1 contained not only barium and nickel in similar low concentrations, but a low level of cadmium was detected. Shibboleth Branch #3 surface water samples again contained elevated concentrations of barium, lead, and nickel compared to the controls. Whereas the barium concentration of Shibboleth Branch #1 was half that of the upstream station, nickel, and zinc was present in similar levels in the Shibboleth Branch samples. None of the dissolved metals exceeded WQSs (MDNR 2010e) in the spring.

Dissolved metals were detected above control concentrations in surface water samples at Trib. Mineral Fork in the spring (Table 13). Trib. Mineral Fork #2 contained barium, cadmium, lead, and nickel. Trib. Mineral Fork #1 contained barium, nickel, and zinc above control concentrations. None of the dissolved metals exceeded WQSs (MDNR 2010e) in the spring.

3.3.2 Pore Water

Several metals of interest were detected in the peepers (pore water samples) in the fall three week sample period (Table 14). Copper, nickel, and zinc were detected in the field blank; subsequently, sample results for these analytes were disregarded from consideration. Barium was found in all tributaries in concentrations at least as high as the surface water samples. Several metals were found in the Mill Creek and Trib. Mineral Fork pore water samples in the fall.

Results from the Mill Creek pore water samples were mixed (Table 14). As noted in Table 14, Pond Creek #2 peepers were missing from the deployment location. Of the metals analyzed, only barium was present in elevated concentrations (654 $\mu\text{g/L}$) in Pond Creek #1 pore water samples. Shibboleth Branch #3 had the highest barium concentration (2610 $\mu\text{g/L}$), elevated cobalt (1.50 $\mu\text{g/L}$), and a hardness corrected lead concentration (41.2 $\mu\text{g/L}$) that exceeded the WQS's chronic exposure level (4 $\mu\text{g/L}$) by a factor of ten. By comparison, the Shibboleth Branch #1 pore water sample contained barium that was approximately three times lower (870 $\mu\text{g/L}$) than upstream, and lead was detected (0.26 $\mu\text{g/L}$) in the pore water sample.

Several dissolved metals also were detected in the pore water samples of Trib. Mineral Fork (Table 14). Trib. Mineral Fork #2 had elevated barium (1070 $\mu\text{g/L}$), cobalt (5.52 $\mu\text{g/L}$), and lead (0.67 $\mu\text{g/L}$). Trib. Mineral Fork #1 contained barium (566 $\mu\text{g/L}$) at about half that of upstream.

3.4 Fine Sediment Percent Coverage and Character

Fine sediment relative percent coverage and character results are presented for Mill Creek tributaries and Trib. Mineral Fork stations for fall 2010. Kruskal-Wallis Analysis of Variants (ANOVA) was used to compare the percent fine sediment between stations and controls. The character of total recoverable barium, cadmium, lead, and zinc was determined and compared to PECs (MacDonald 2000) to account for individual metals levels. The potential risk due to a mixture, or combination of metals was examined using ΣPEQ and mean PEQ thresholds (MacDonald et al. 2009). The threshold levels (MacDonald et al. 2009) for the ΣPEQ cadmium, lead, zinc mixture is 7.92, whereas the mean PEQ is 1.11.

3.4.1 Fine Sediment Percent Coverage

Fine sediment relative coverage for Mill Creek stations was examined in the 2008-2009 study (MDNR 2009a), but is reiterated here (Table 15; Appendix C). In that study, Pond Creek #2 and #1 had significantly higher ($p < 0.05$) coverage of fine sediment than the controls. Shibboleth Branch #3 was not significantly different ($p > 0.05$) from the controls; however, fine sediment was present in a patchy distribution (MDNR 2009a). Shibboleth Branch #1 had significantly higher ($p < 0.05$) coverage than the controls.

The fine sediment relative percent coverage at Trib. Mineral Fork stations was examined during this study (Table 15; Appendix C). Trib. Mineral Fork #2 percent coverage was significantly higher ($p < 0.05$) than the controls (Table 15; Appendix C). The percent

coverage was 75.5% (± 21.9) at Trib. Mineral Fork #2, as opposed to 19.2 for the mean of controls. Trib. Mineral Fork #1 percent coverage was also significantly higher ($p < 0.05$) than the controls (Table 15; Appendix C). The percent coverage was 42.1 (± 34.1) compared to 19.2 for the mean of controls.

3.4.2 Fine Sediment Character

Fine sediment character results from the previous study (MDNR 2009a) were included and are compared to individual metals thresholds and mixture of metals thresholds (Table 16; Table 17). PECs (MacDonald et al. 2000) and PEQs (Besser et al. 2009a) were compared to individual metals concentrations in Mill Creek tributaries and Trib. Mineral Fork stations (Table 16). To account for the mixture of metals (cadmium, lead, zinc), Σ PEQ or mean PEQ thresholds (MacDonald et al. 2009) were compared to levels found in Mill Creek and Trib. Mineral Fork stations (Table 17).

In the Mill Creek watershed, both Pond Creek stations exceeded the PEC for zinc. Shibboleth Branch #3 exceeded the PECs for lead and zinc. Shibboleth Branch #1 exceeded the PECs for cadmium, lead, and zinc. Each metal that exceeded PECs, also exceeded the acceptable PEQ.

The influences attributed by a mixture or combination of metals (cadmium, lead, zinc) were examined for Mill Creek stations and are presented as Σ PEQ and mean PEQ (Table 17). Pond Creek stations did not exceed the Σ PEQ threshold (7.92) or the mean PEQ threshold (1.11) for the three metals. Shibboleth Branch #3 exceeded both Σ PEQ and mean PEQ threshold levels for the cadmium, lead, zinc combination. Shibboleth Branch #1 was near, but did not exceed the Σ PEQ threshold; however, the mean mixture of metals threshold was exceeded by over two-fold.

Total recoverable barium, lead, and zinc each were present in Trib. Mineral Fork sediment samples in concentrations above their individual PECs (Table 16). Trib. Mineral Fork #2 fine sediment contained lead (329 mg/kg) over twice the PEC (128 mg/kg), and zinc (525 mg/kg) above the PEC (459 mg/kg). The fine sediment at Trib. Mineral Fork #1 contained total recoverable lead (521 mg/kg) well above the PEC (128 mg/kg). Each metal that exceeded PECs, also exceeded the acceptable PEQ.

The influences attributable to a mixture or combination of metals (cadmium, lead, zinc), was examined for Trib. Mineral Fork stations using Σ PEQ and mean PEQ threshold levels (MacDonald et al. 2009; Table 17). The Trib. Mineral Fork #2 Σ PEQ (4.0) did not exceed the Σ PEQ threshold (7.92); however, the mean PEQ (1.33) was higher than the threshold (1.11). Trib. Mineral Fork #1 fine sediment Σ PEQ (5.3) did not surpass the threshold; however, the mean PEQ (1.77) exceeded the mean threshold (1.11).

Table 12
Surface Water (Grab sample) Dissolved Metals and Hardness for Tributaries and Controls, Fall 2010

Parameter Station	Ba	Cd	Ca	Co	Cu	Pb	Mg	Ni	Zn	HARD CaCO ₃
Pond Cr. #2	509	<0.20	34.8	<1.0	0.53	<0.25	20.0	0.33	3.92	169
Pond Cr. #1	680	<0.20	46.7	<1.0	0.69	<0.25	28.8	0.44	4.97	235
Shibboleth Br. #3	1610	<0.20	35.1	<1.0	0.62	0.27	21.0	0.36	3.32	174
Shibboleth Br. #1	776	<0.20	47.7	<1.0	0.68	<0.25	30.3	0.48	4.80	244
Trib. Mineral Fork #2	505	<0.20	57.7	<1.0	1.17	<0.25	37.8	0.97	17.5	300
Trib. Mineral Fork #1	544	<0.20	47.6	<1.0	0.82	<0.25	34.2	0.85	2.84	260
Brazil Cr. #1 c	86.1	<0.20	27.9	<1.0	0.70	<0.25	16.3	<0.25	2.28	137
Courtois Cr. #1A c	45.8	<0.20	28.7	<1.0	0.59	<0.25	17.3	<0.25	1.50	143
Courtois Cr. #1B c	45.1	<0.20	28.7	<1.0	0.51	<0.25	17.3	<0.25	1.34	143
E Fk Huzzah Cr. #1 c	48.9	<0.20	36.4	<1.0	0.75	<0.25	22.4	<0.25	8.13	183
W Fk Huzzah Cr #1 c	38.8	<0.20	29.3	<1.0	0.52	<0.25	17.6	<0.25	1.36	146
Shoal Cr. #1 c	51.0	<0.20	42.9	<1.0	1.81	<0.25	25.8	<0.25	3.34	213

Units µg/L; **Bold**=notable; c=controls results from fall 2008 (MDNR 2009a).

Table 13
Surface Water (Grab sample) Dissolved Metals and Hardness for Tributaries and Controls, Spring 2011

Parameter Station	Ba	Cd	Ca	Co	Cu	Pb	Mg	Ni	Zn	HARD CaCO ₃
Pond Cr. #2	570	<0.02	18.8	<1.0	1.08	<0.25	11.6	0.79	5.33	94.7
Pond Cr. #1	498	0.02	27.2	<1.0	1.08	<0.25	16.4	0.80	6.50	135
Shibboleth Br. #3	1190	<0.02	26.3	<1.0	1.05	0.29	16.4	0.71	7.18	133
Shibboleth Br. #1	679	<0.02	36.2	<1.0	1.23	<0.25	21.6	0.87	10.8	179
Trib.Mineral Fork #2	375	0.03	44.6	<1.0	2.21	0.36	27.5	1.30	18.7	225
Trib.Mineral Fork #1	392	<0.02	44.8	<1.0	1.77	<0.25	27.3	1.16	9.64	224
Brazil Cr. #1A c	75.9	<0.20	23.9	<1.0	1.00	<0.25	13.8	0.30	7.77	116
Brazil Cr. #1B c	70.7	<0.20	23.7	<1.0	0.64	<0.25	13.7	<0.25	2.21	116
Courtois Cr. #1 c	28.6	<0.20	17.2	<1.0	0.92	<0.25	9.91	<0.25	2.28	83.7
E Fk Huzzah Cr. #1 c	28.5	<0.20	30.4	<1.0	0.71	<0.25	18.7	<0.25	6.82	153
W Fk Huzzah Cr. #1 c	32.7	<0.20	24.2	<1.0	0.54	<0.25	14.6	<0.25	1.65	121
Shoal Cr. #1 c	38.7	<0.20	34.6	<1.0	1.23	<0.25	20.5	0.29	2.09	171

Units µg/L; **Bold**=notable; c=controls results from spring 2009 (MDNR 2009a).

Table 14
Pore Water (Peeper samples) Dissolved Metals and Hardness for Tributaries Fall 2010

Parameter Station	Sample Number	Ba	Cd	Ca	Co	Cu	Pb	Mg	Ni	Zn	HARD CaCO ₃
Pond Cr. #2	*	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
Pond Cr. #1	1006945	654	<0.2	46.6	<1.0	1.20	<0.25	29.4	1.36	6.45	237
Shibboleth Br. #3	1006939	2610	<0.2	34.6	1.50	1.35	41.2 c	20.8	1.51	64.5	172
Shibboleth Br. #1	1006938	870	<0.2	49.0	<1.0	0.58	0.26	30.7	1.36	10.4	249
Trib. Mineral Fork #2	1006943	1070	<0.2	64.5	5.52	0.31	0.67	45.0	2.52	24.7	346
Trib. Mineral Fork #1	1006942	566	<0.2	55.7	<1.0	3.63	<0.25	41.8	2.49	15.2	311
Field Blank	1006940	1.13	<0.2	<0.10	<1.0	15.1	<0.25	<0.10	2.00	40.1	1.65

Units µg/L; *Peeper lost in stream; Bold=elevated or interesting result; c=above chronic WQS

Table 15

Fine Sediment Percent Coverage by Station, Grid, and Transect. Mean, Standard deviation, and Significance Level ($p < 0.05$) Using Kruskal-Wallis One-Way Analysis of Variance on Ranks (Analyses in Appendix C).

Grid- Transect	PC #2	PC #1	SB #3	SB #1	TMF #2	TMF #1	BC #1	CC #1	EFHC #1	WFHC #1	SC #1
1-1	92	25	6	40	35	15	30	27	3	3	3
1-2	99	55	3	43	65	3	15	17	4	3	1
1-3	95	10	90	15	23	15	7	7	7	3	1
1-4	95	40	25	17	77	75	40	10	9	7	1
1-5	90	5	7	33	71	87	23	4	1	7	7
1-6	85	50	13	20	89	15	23	7	4	1	1
2-1	98	27	5	77	97	3	9	3	4	4	23
2-2	95	45	6	13	97	65	5	1	3	3	20
2-3	95	10	5	23	97	10	80	10	1	2	23
2-4	95	25	85	10	97	3	35	17	5	3	87
2-5	90	55	5	7	97	35	40	5	1	70	80
2-6	89	13	3	15	97	10	35	1	5	53	63
3-1	95	75	90	43	73	87	2	35	45	3	3
3-2	95	23	17	77	73	89	9	12	45	1	15
3-3	87	70	87	80	73	90	1	70	17	5	7
3-4	97	93	87	85	83	55	23	25	17	7	20
3-5	90	27	95	77	60	65	15	1	7	2	8
3-6	97	23	95	67	55	35	7	13	13	3	13
MEAN	93.3	37.3	40.2	41.2	75.5	42.1	22.2	14.7	10.6	10.0	20.9
S.D.	3.9	24.8	41.1	28.4	21.9	34.1	19.5	16.8	13.4	19.1	27.1
KW ANOVA	p<0.05	p<0.05	NS	p<0.05	p<0.05	p<0.05	19.2 Mean of controls				

Key: Test Stations= PC-Pond Creek (2009); SB-Shibboleth Branch (2009); TMF-Trib. Mineral Fork (2010), Controls (2009)=BC-Brazil Creek, CC-Courtois Creek, EFHC-East Fork Huzzah Creek, WFHC-West Fork Huzzah Creek, SC-Shoal Creek. All results, except Trib. Mineral Fork, are taken from 2009 report (MDNR 2009a)

Table 16
Total Recoverable Metals Character in the Fine Sediment (<2.0mm): Barium, Cadmium, Lead, and Zinc Concentrations (mg/kg Dry Weight)

Parameter Station	Barium	Cadmium	Lead	Zinc
Pond Creek #2 *	1580	0.683	46.6	488
Pond Creek #1 *	1460	0.594	96.8	525
Shibboleth Branch #3 *	2890	0.638	836	697
Shibboleth Branch #1*	428	9.52	607	553
Trib. Mineral Fork #2	1640	1.43	329	525
Trib. Mineral Fork #1	2400	0.435	521	398
Brazil Creek #1 *	24.8	0.101	49.3	54.6
Courtois Creek #1*	13.3	0.100	8.7	9.5
E Fk Huzzah Cr.#1a*	19.0	0.599	15.1	64.5
E Fk Huzzah Cr.#1b*	18.6	0.381	13.4	45.6
W Fk Huzzah Cr.#1 *	21.6	0.100	10.8	9.5
Shoal Creek #1*	15.7	0.169	15.9	45.4
PEC	--	4.98 mg/kg	128 mg/kg	459 mg/kg

PEC=Probable Effects Concentration (MacDonald et al. 2000); a and b=duplicate; light gray=candidate reference stations; **Bold**=above PEC; *=taken from Fall 2008 results (MDNR 2009a) for comparison.

Table 17
Probable Effects Quotients (PEQ) and Mixture of Metals (Σ PEQ and mean PEQ) Threshold Levels (Besser et al. 2009a; MacDonald et al. 2009) for Total Recoverable Metals in the Tributaries

Parameter PEQ Station	Cadmium	Lead	Zinc	Σ PEQ	Mean PEQ
Pond Creek #2	0.137	0.364	1.063	1.564	0.521
Pond Creek #1	0.119	0.756	1.144	2.019	0.673
Shibboleth Branch #3 *	0.128	6.531	1.519	8.178	2.726
Shibboleth Branch #1	1.912	4.742	1.205	7.859	2.620
Trib. Mineral Fork #2	0.287	2.570	1.144	4.001	1.334
Trib. Mineral Fork #1	0.087	4.070	1.153	5.310	1.770
Thresholds PEQ	>1.0	>1.0	>1.0	ΣPEQ>7.92	Mean PEQ>1.11

Bold station=station intermittently or *continuously partially supporting the AQL; **Bold metric**=above threshold PEQs (Besser et al. 2009a), Σ PEQ (MacDonald et al. 2009), and mean PEQ (MacDonald et al. 2009).

4.0 Discussion

The discussion is grouped by Mill Creek Tributaries and Tributary of Mineral Fork, including both seasons. Major sections within each group include: stream habitat assessments, macroinvertebrate community assessments, general water quality, dissolved metals (surface and pore water), as well as fine sediment (coverage and character). Results may be compared with the same Mill Creek tributaries that were included in the earlier MDNR study (MDNR 2009a).

4.1 Mill Creek Tributaries

Mill Creek tributaries included in this study are Pond Creek (WBIDs 2128 and 2127) and Shibboleth Branch (WBIDs 2120 and 2119). Two stations were assigned to each stream in the same locations as the 2008-2009 study (MDNR 2009a).

4.1.1 Stream Habitat Assessment

Stream habitat assessments were conducted for Mill Creek tributaries during the earlier MDNR study (MDNR 2009a), but are reiterated in this report for consistency. Generally, Pond Creek #2 contained mostly bedrock substrate and this reduced available habitat may have been partially responsible for intermittent biological impairment. Runoff from the nearby gravel road appeared to contribute fine sediment to the substrate bedload. The Pond Creek #1 substrate was more heterogeneous in size-classes, and this station had fully supporting MSCI scores during both sample seasons.

The substrate at Shibboleth Branch #3 was assessed twice in the earlier study (MDNR 2009a): once using the sediment estimation and characterization procedure, and the second time was during a SHAPP. Benthic sediment coverage was much higher when the SHAPP was conducted than was estimated when the fine sediment sampling occurred. This difference suggested that fine sediment coverage fluctuated and may have contributed to consistently low MSCI scores in earlier studies. Shibboleth Branch #1 had a high percentage of bedrock as its substrate, and a very apparent braided sediment bar near the downstream reach of the station. This sediment bar may have been deposited as a result of an earlier barite dam failure (Duchrow 1978), or possibly from other sources such as persistent or periodic runoff. Station #1 was consistently fully supporting during the earlier study irrespective of the bar (MDNR 2009a).

4.1.2 Macroinvertebrate Community

Pond Creek #2 was fully supporting in the fall, but partially supporting of the AQL beneficial use category in the spring. Station #2 was consistently impaired during the earlier study (MDNR 2009a). This station maintained a consistently high BI during this and the earlier study, which suggests that the macroinvertebrate community assemblage is more tolerant to organic or nutrient input. In fact, the percent sensitive taxa distribution showed 45 to 50 percent of the sample had a BI of ≥ 7.5 , which is over two times higher than the BIOREF BI for that EDU. The Pond Creek #2 MSCI scores did not change during either season when compared to similar size control criteria, although the score did change during spring in the earlier study (MDNR 2009a). This change suggests

that stream size did not obviously affect the MSCI scores this time, and the stream was sometimes comparable to a high quality smaller stream.

Pond Creek #1 was fully supporting of the AQL during both seasons. Station #1 had consistently fully supporting scores in the 2009 study as well. The BI score was consistently low using BIOREF criteria, suggesting that organic influences were not influential at this station. However, when metrics were compared to similar size control criteria, the BI score was less than optimal. This is also consistent with the 2009 findings (MDNR 2009a). Although Pond Creek #1 was fully supporting compared to BIOREF criteria, the change in the BI score when compared to similar size control criteria suggests that the macroinvertebrate community is composed of organisms that are more tolerant than high quality small control streams.

Shibboleth Branch #3 was partially supporting of the AQL beneficial use in both fall 2010 and spring 2011 seasons. This station was partially supporting during both seasons in the previous study as well (MDNR 2009a), which suggests that the station is consistently impaired. The TR, EPTT, and number of mayfly taxa were much lower at Shibboleth Branch #3 compared to the downstream Shibboleth station, the controls, and any of the other tributaries in this study. Although the macroinvertebrate community was consistently dominated by Chironomidae, Shibboleth Branch #3 had consistently low BI values in this and earlier studies (MDNR 2009a). However, the BI was suboptimum when compared to similar sized control criteria. This difference in BI threshold values suggests that Shibboleth Branch #3 has taxa that are similarly tolerant to larger BIOREF streams, but more tolerant than high quality small control streams. Based on BI values, the Shibboleth Branch #3 macroinvertebrate community does not show any obvious signs of organic pollutant effects.

Shibboleth Branch #1 was fully supporting in the fall and partially supporting the AQL beneficial use in the spring 2011. The fall sample had a suboptimal BI score, whereas all other metrics were optimal. Metrics that contributed to partial support in the spring included low EPTT, high BI, and low SDI scores. During this study, the BI score was consistently suboptimal at this station, which is similar to results in the earlier study (MDNR 2009a). The percent sensitive taxa distribution showed that over 30 percent of the community had a BI above 7.5, whereas about 20 percent of the community had a BI above 7.5 at BIOREF stations. This elevated BI suggests that the Shibboleth Branch #1 community composition is more tolerant to organic influences and disturbance than a BIOREF stream. Despite having consistently elevated BI values, spring was the first time in four sample seasons that impairment was calculated. Interestingly, when Shibboleth Branch #1 was assessed using control stream criteria, the original MSCI score changed from the partially supporting category to full support of the AQL. This difference suggested that the station was more similar to a smaller high quality stream; however, with an elevated BI, the macroinvertebrate community was still more tolerant in Shibboleth Branch #1 than a high quality small control stream community.

Interestingly, all of the Mill Creek tributaries had suboptimum BI scores during both seasons when they were compared to control stream criteria. This suggests that all of the tributaries had more tolerant communities than high quality small streams, whether or not they were fully supporting of the AQL.

4.1.3 General Water Quality

The water quality parameters that were included did not indicate that an obvious pollution influence existed at Mill Creek tributaries. However, low levels of total nitrogen, nitrate+nitrite-N, and chloride were present during both seasons at Pond Creek #2 and Shibboleth Branch #1, which may indicate that these streams receive organic influences. These indicators were consistently observed at these two stations in the 2009 study (MDNR 2009a) as well, which suggests that the influence is persistent. BIs were consistently high during this and the earlier study, which also indicates that organic influences may affect the macroinvertebrate community composition at Pond Creek #2 and Shibboleth Branch #1. All surface water quality results, however, were within WQSS (MDNR 2010e) during sampling.

4.1.4 Dissolved Metals

Dissolved cadmium, lead, nickel, and zinc were detected in surface water and pore water samples collected in Mill Creek tributaries. These findings are similar to the previous study (MDNR 2009a), as well as mine-related studies by Besser et al. (2009b) in Big River, Brumbaugh et al. (2007) in the New Lead Belt (southeastern) Missouri, and Allert et al. (2008, 2009, 2011), also in the New Lead Belt and in Southwest Missouri. The metals and concentrations discussed here are elevated compared to the controls, but are not necessarily above limits of the WQSS (MDNR 2010e) unless specifically noted.

4.1.4.1 Surface Water

The surface water samples from Pond Creek stations consistently contained dissolved metals. Barium and nickel concentrations were elevated compared to the controls at both stations and during both seasons. In addition, cadmium was detected at station #1 in the spring. These metals were detected in very low concentrations and none exceeded WQSS, which suggests that dissolved metals in the surface water were not obviously affecting the macroinvertebrate communities. This is consistent with earlier findings at Pond Creek stations (MDNR 2009a).

Shibboleth Branch stations contained dissolved metals in the surface water that followed interesting trends from this and the earlier study. Shibboleth Branch #3 contained the highest barium concentrations of all Mill Creek tributaries and the controls, along with consistently elevated lead and nickel. Lead concentrations fluctuated in the earlier study (MDNR 2009a). However, when lead was detected, it was found in higher concentrations among upstream stations when compared to downstream. Again, dissolved lead was not detected in the surface water at station #1. Dissolved barium and nickel were consistently detected at station #1, and zinc was detected in the spring. Interestingly, the barium concentration at station #1 was approximately half of the

concentration found upstream. The slight trends in dissolved lead and barium concentrations from upstream (high) to downstream (low) in this study and the earlier study suggests that a source for these metals is at or upstream of station #3. This also suggests that metals concentrations in the surface water fluctuate given suitable conditions. Although these metals were present in the surface water during sampling, none exceeded WQSs (MDNR 2010e) and were not obvious contributors to impairment at station #3.

4.1.4.2 Pore Water

Several dissolved metals were detected in pore water samples that were collected in Mill Creek tributaries, which is consistent with previous work by Besser et al. (2009b) in Big River pore water, and Brumbaugh et al. (2007) in mine-related streams of the New Lead Belt Viburnum Trend. Usually pore water concentrations were similar to surface water samples; however, there were differences at some stations, which is similar to the findings of Brumbaugh et al. (2007). Copper, nickel, and zinc results were not included in the pore water section due to the presence of these metals in the field blank.

As mentioned earlier, Pond Creek #2 peepers were missing when the samplers were to be retrieved. So, the Pond Creek #1 peeper sample was used to characterize the pore water metals in that tributary. When the pore water sample was compared to the controls, the barium concentration was elevated. No other metal analyzed was detected above control concentrations. Dissolved metals in pore water did not exceed WQSs (MDNR 2010e), and concentrations were similar to the corresponding surface water sample.

Interestingly, the Shibboleth Branch #3 peeper sample contained a dissolved lead concentration that was ten times higher than the WQS (MDNR 2010e). Subsequently, the PWTU was then over ten and the Σ PWTU for cadmium, lead, and zinc would be well over ten, which was much higher than the Σ PWTU threshold of 1.03 (MacDonald et al. 2009). The high mixture quotients were due to the extremely high lead concentration in the pore water sample. This station was partially supporting of the AQL beneficial use during every visit in this and the earlier study (MDNR 2009a). Poulton et al. (2009) found that concentrations of metals in sediment pore water were highly significantly correlated with biotic condition scores, and that dissolved lead was among the important metals in pore water that may contribute to lower scores. Pore water metals may be affecting the macroinvertebrate community at station #3, as this station has been consistently partially supporting of the AQL. Pore water peepers were not deployed at Shibboleth Branch #3 in the spring, so it is not known whether pore water metals again led to this station's impaired status.

It is also interesting to note that dissolved lead in the fall pore water sample was over 150 times higher than the fall surface water sample. This observation suggests that concentrations are higher in the substrate interstices; or that a high lead concentration event may have occurred in the system at some point after surface water was collected and before the peepers were retrieved; and that lead concentrations may fluctuate greatly.

Peepers may take a few days to normalize with the ambient dissolved metals concentrations (Brumbaugh et al. 2007). Thus, the peepers will effuse higher concentrations over a period of days, which may explain how the peeper captured a higher concentration spike.

As mentioned, Poulton et al. (2009) found that dissolved lead in pore water was one of the most important parameters for categorizing sites in their study of the Viburnum Trend. Taxa richness and EPTT are the best biological indicators of these effects (Soucek et al. 2000; Clements et al. 2000). Mayflies and stoneflies are among the most sensitive macroinvertebrate groups to heavy metals contamination in streams (Ryck 1974; Burrows and Whitton 1983; Kiffney and Clements 1994; Carlisle and Clements 1999; Yuan and Norton 2003; Poulton et al. 2009), and their tolerance may be pH dependent (Feldmann and Connor 1992; Yuan and Norton 2003; Poulton et al. 2009). The TR, EPTT, and number of mayflies were much lower in Shibboleth Branch #3 during both seasons (Appendix A), while pore water lead was very high in the fall. Certain mayflies such as *Caenis* spp. and *Maccaffertium mediopunctatum* were much less abundant or absent at station #3 than at station #1 during both seasons, and the earlier study (MDNR 2009a). The shift in community structure and coinciding increased pore water metal indicated that metals may be affecting the macroinvertebrate community at Shibboleth Branch #3.

Interestingly, Heptageniidae, which is generally thought to be sensitive to metals, was among the dominant families at Shibboleth Branch #3. This seems to contradict the suggestion that impairment may be due to excessive metals concentrations. However, *Maccaffertium pulchellum* was the taxon that made Heptageniidae one of the dominant families at station #3 in both fall and spring samples. Furthermore, *M. pulchellum* was much more abundant at Shibboleth Branch #3 compared to the downstream station Shibboleth Branch #1 and the controls in this and the earlier study as well. Not only was *M. pulchellum* apparently unaffected by the conditions of station #3, it appeared to be able to exploit them. If excessive heavy metals were the condition that affected the macroinvertebrate community at this station, *M. pulchellum* exhibited a greater tolerance than other members of the same mayfly family. For example, *M. mediopunctatum* was not found in station #3, yet it was found in all Shibboleth Branch stations downstream during this and the earlier study. This observation suggests taxa within the same family have varying tolerance levels to stressors. Both the increase in one genus or species and decrease in another, suggests that family level generalizations regarding this adverse condition are not accurate.

Dissolved lead was detected in the pore water at Shibboleth Branch #1, although in a much lower concentration than upstream. Dissolved lead was not detected in the fall surface water sample suggesting that concentrations were slightly higher in the substrate than the water column. Brumbaugh et al. (2007) found dissolved metal concentrations to be slightly higher in peepers than surface water grab samples. The pore water dissolved lead concentration was low in the fall at station #1 when the station was fully supporting the AQL. Pore water was not sampled in the spring when station #1 was partially

supporting, so the low MSCI score could not be associated with dissolved metals concentrations in the pore water.

4.1.5 Fine Sediment Coverage

Fine sediment coverage was examined in the earlier study of Mill Creek tributaries (MDNR 2009a). Several Pond Creek and Shibboleth Branch stations had fine sediment coverage that were significantly greater ($p < 0.05$) than the controls.

Both Pond Creek stations had significantly higher ($p < 0.05$) fine sediment coverage than the controls. Pond Creek #2 had a mean coverage of over 90 percent in the earlier study and was impaired during both seasons. It was suggested in the earlier study (MDNR 2009a) that Pond Creek #2 impairment may have been associated with fine sediment or with poor habitat. This study revealed that Pond Creek #2 was again partially supporting of the AQL in the spring. As mentioned in the SHAPP section (4.1.1), runoff from the gravel road adjacent to the stream may have been contributing to the substrate coverage. Based on the difference in coverage between controls and test stations, it is possible that the Pond Creek #2 community was affected by fine sediment.

Taxa intolerant of excessive fine sediment (Zweig and Rabeni 2001) such as *Caenis* spp. and *Maccaffertium* (*Stenonema*) spp. were among the dominant taxa at Pond Creek stations. *M. pulchellum*, which is considered to be very intolerant of fine sediment, was one of the most prevalent taxa; and its numbers made Heptageniidae one of the dominant families at Pond Creek. However, another Heptageniidae, *M. mediopunctatum* was not present upstream in Pond Creek #2, but was observed downstream in station #1 during both seasons. The presence of a sensitive taxon and absence of another within a generally sensitive family did not clearly identify fine sediment as a negative influence. However, invertebrates may respond differently to various proportions of sand and silt (Zweig and Rabeni 2001), which may explain why some sensitive taxa were present and some sensitive taxa were absent. It appears that at least one taxon within the family thrived, while another appeared to be influenced by a stressor.

The mean fine sediment percent coverage at Shibboleth Branch #3 was approximately 40 percent, which was not significantly greater than the controls. However, fine sediment coverage may fluctuate significantly, as was shown by previous SHAPP observations (MDNR 2009a), by a relative percent coverage standard deviation of 41 percent. Fine sediment fluctuations may have contributed to consistent impairment at Shibboleth Branch #3.

Fine sediment intolerant taxa such as *Caenis* spp., *Isonychia* spp., and *M. mediopunctatum* (Zweig and Rabeni 2001) were much less abundant upstream than downstream, which suggests that fine sediment may be contributing to impairment. However, *Maccaffertium pulchellum* was abundant and is also considered intolerant. If fine sediment is affecting the stream, it may be that *M. pulchellum* may not be as intolerant as the aforementioned mayflies; family level tolerance generalizations

regarding tolerance to fine sediment may not be accurate and fine sediment may have contributed to impairment at station #3.

The mean fine sediment coverage at Shibboleth Branch #1 was approximately 41 percent and was significantly greater ($p < 0.05$) than the controls. The difference in fine sediment coverage between controls and this test station suggests that fine sediment may have contributed to the biological impairment in the spring. However, this station contained many more TR and EPTT than the upstream, which are considered to be negatively correlated with increasing fine sediment coverage (Zweig and Rabeni 2001). Fine sediment intolerant taxa such as *Caenis* spp. were abundant and *M. mediopunctatum* was present, which suggests that fine sediment coverage may not be a contributor to impairment at station #1 in the spring.

4.1.6 Fine Sediment Character

The fine sediment in the substrate was characterized for cadmium, lead, and zinc content. Mill Creek tributaries substrates were sampled in the 2008-2009 MDNR study (MDNR 2009a). Those results are compared to additional threshold values that were not included in the earlier MDNR study. Individual metals thresholds (PEC, MacDonald et al. 2000; PEQ, Besser et al. 2009a), and mixture of metals thresholds (Σ PEQ and mean PEQ, MacDonald et al. 2009) were compared to this data.

The substrate's fine sediment at both Pond Creek stations contained zinc that exceeded the appropriate PEC in the 2008-2009 MDNR study (MDNR 2009a). The PEQ slightly exceeded one, which suggests that the metal concentration may increase the probability of toxic effects on the macroinvertebrate community (Besser et al. 2009a). Neither threshold was exceeded for the mixture of metals (cadmium, lead, zinc).

Individual heavy metals made up a portion of the fine sediment at Shibboleth Branch stations in the 2008-2009 study. At Shibboleth Branch #3 the lead concentration was over six times higher than the PEC, and zinc was one and a half times higher than the PEC. Shibboleth Branch #1 contained cadmium, lead, and zinc above the PECs in the sediment. Each PEQ threshold was exceeded at station #1. It is possible that the individual metals in the fine sediment increase the probability of toxic effects on the community, as mentioned by Besser et al. (2009a). Heavy metals may be contributing to the partial support status of the aquatic life beneficial use category at these stations.

The Σ PEQ and mean PEQs were compared to their respective threshold limits to assess the effects of a mixture of metals (MacDonald et al. 2009) at Shibboleth Branch stations. The mixture or combination of cadmium, lead, and zinc exceeded both of these thresholds at Shibboleth Branch #3. The Shibboleth Branch #1 combined metals concentration did not exceed the Σ PEQ, but the mean PEQ was exceeded. A mixture of metals exceeding threshold limits is likely to be toxic to benthic macroinvertebrates (MacDonald et al. 2009). Shibboleth Branch #3 and #1 may have been impaired by a mixture of heavy metals in the fine sediment.

4.2 Tributary of Mineral Fork

This was the first study conducted by ESP of Tributary of Mineral Fork (WBID 2115). Two stations were allocated for this project, which included a stream habitat assessment, macroinvertebrate community and water quality analyses, dissolved metals (surface and pore water), and fine sediment (coverage and character) analyses. Since this is the first study, no temporal comparisons can be made.

4.2.1 Stream Habitat Assessment

The SHAPP scores were similar from station to station and to the controls at Trib. Mineral Fork stations. Both stations were above the 75th percent similarity criterion of SHAPP controls. Stream habitat quality should not affect the results of this study.

4.2.2 Macroinvertebrate Community

Trib. Mineral Fork #2 was fully supporting of the AQL beneficial use in the fall and the spring. It had consistently high BI values with over 40 percent of individuals in the sample having a BI above 7.5. Elevated BI values suggest that organic pollutant influences may be present, although not to the point of impairing the station's macroinvertebrate community. Interestingly, the Mineral Fork #2 MSCI score did not change when metrics were compared to similar size control criteria, so stream size did not affect the overall score. The BI remained consistently high when metrics were compared to control criteria, which suggests that the Trib. Mineral Fork #2 macroinvertebrate community was more tolerant to organic influence or disturbance than would be found in the high quality small streams.

Trib. Mineral Fork #1 was fully supporting in the fall of 2010 and partially supporting in the spring of 2011. TR, EPTT, and SDI contributed to the low MSCI score in the spring. This suggests that the community was smaller, less sensitive, and less diverse than a BIOREF stream. The BI was consistently low when compared to the BIOREF criteria, which suggests that organic pollutants were not an obvious or substantial factor affecting community in this station. Additionally, when station #1 was compared to similar-size control criteria using spring data, the TR, EPTT, BI, and SDI scores increased and the MSCI changed to fully supporting. This change suggests that the station is more similar to the control streams and that stream size may be a factor in determining the MSCI score at this site. However, the change in BI to less than optimum suggests that taxa at station #1 were generally more tolerant than are assembled in a high quality small control stream.

Interestingly, each of the Trib. Mineral Fork stations had elevated BI metric scores when compared to similar-size control stream criteria. This metric suggests that the Trib. Mineral Fork macroinvertebrate community is generally more tolerant to organic influences or disturbance than similarly sized control streams.

4.2.3 General Water Quality

Water quality parameters were not exceptional; however, organic or nutrient indicators were apparent at Trib. Mineral Fork. Consistently elevated (i.e. compared to controls) conductivity, low levels of total nitrogen, nitrate+nitrite-N and chloride at Trib. Mineral Fork #2 are suggestive of a persistent organic input. A consistently high BI suggests that the macroinvertebrate community was also more tolerant to organic influences or disturbance than BIOREF stations. It is apparent that some organic influence is potentially available upstream, such as houses, yards, etc. In addition to possible domestic influences, a barite tailings pond is located upstream of station #2. These water quality parameters are commonly elevated in mine related streams (Poulton et al. 2009; Allert et al. 2011). All parameters were within WQSs (MDNR 2010e).

Water quality parameters did not provide an obvious explanation of why Trib. Mineral Fork #1 was impaired in the spring. However indicators such as conductivity, total nitrogen, nitrate+nitrite-N and chloride were elevated relative to controls in the fall, whereas total conductivity, nitrogen and chloride were slightly elevated in the spring. Consistently optimum BIs suggest that organic influences may not have been an obvious contributor to impairment in the spring. It appears that the potential upstream (#2) organic influence does not extend downstream to Trib. Mineral Fork #1. This also suggests that another influence may have affected the community. Water quality parameters, such as high conductivity, total nitrogen (Poulton et al. 2009; Allert et al. 2011), and chloride in low concentrations also are common in mine related streams. Station #1 may be affected by mine related influences or, as shown earlier, by the size of the stream. It may be that organic influences were present, but were not obviously responsible for the partially supporting spring MSCI score. All parameters were within WQSs (MDNR 2010e).

Compared to controls and other test stations, conductivity was the highest at both Trib. Mineral Fork stations. Conductivity is commonly elevated in mine related streams of the Midwest (Allert et al. 2011). Allert et al. (2011) and Poulton et al. (2009) found elevated conductivity downstream from mine related sites in the Viburnum Trend. Furthermore, Poulton et al. (2009) found that conductivity was correlated with biotic condition score. Both Trib. Mineral Fork stations are downstream from a tailings pond and the stations bracket a smelter pond, which may have been responsible for elevated conductivity.

4.2.4 Dissolved Metals

Dissolved metals were observed in surface water and pore water in Trib. Mineral Fork. Dissolved metals such as barium were elevated, yet were not as high as other mine related streams in this study. As observed by Brumbaugh (2007) both surface and pore water metals concentrations tend to be similar to one another, or pore water is higher. None of the Trib. Mineral Fork dissolved metals in surface water exceeded WQSs (MDNR 2010e) in either sample season.

4.2.4.1 Surface Water

Dissolved metals were consistently elevated in the surface water of Trib. Mineral Fork compared to the control stations. Barium, nickel, and zinc were detected in the both seasons at station #2, whereas cadmium and lead were also detected in the spring. Downstream in station #1 barium and nickel were consistently present, whereas zinc was higher than control concentrations in the spring. Zinc was consistently much higher at station #2, than at station #1. Despite the presence of these metals in the surface water, none exceeded WQSs (MDNR 2010e) and were not obvious contributors to impairment at station #1 in the spring.

4.2.4.2 Pore Water

Dissolved metals were detected in the pore water samples taken from Trib. Mineral Fork. Barium, cobalt, and lead were elevated at Trib. Mineral Fork #2, but barium was the only elevated parameter in pore water at station #1. The concentrations of heavy metals in pore water could not be associated with the partially supporting category at station #1 in the spring because pore water samples were not collected in the spring. Copper, nickel, and zinc results were not included in the pore water section due to the presence of these metals in the field blank.

Interestingly, lead and cobalt were detected in the pore water sample from Trib. Mineral Fork #2, but it was not detected in the corresponding surface water sample. Brumbaugh et al. (2007) observed higher concentrations of heavy metals in some pore water samples than corresponding surface water samples. These results suggest that metals concentrations may be higher in pore water, or that dissolved metals concentrations fluctuated during deployment before the peepers were retrieved. In other samples however, they found the two water samples (pore and surface) to be similar, which illustrated an interaction between the surface and pore water (Brumbaugh et al. 2007). None of the pore water samples at Trib. Mineral Fork exceeded WQSs (MDNR 2010e). Pore water should be sampled using peepers as standard procedure in future mine related stream studies, as well as in mine related streams in where biological assessments and stream studies have already been completed.

As mentioned earlier, elevated metals found in pore water using peepers and not in surface water samples suggests that: 1) dissolved metals concentrations may be higher in the substrate interstices; 2) peepers may be more effective at collecting intermittent influxes of metals than grab samples; 3) pore water should be sampled routinely using peepers; and 4) macroinvertebrate communities may be affected by intermittent heavy metals concentrations that are not collected using grab samples.

4.2.5 Fine Sediment Coverage

Trib. Mineral Fork stations had significantly greater ($p < 0.05$) fine sediment percent coverage than the control streams. Trib. Mineral Fork #2 was consistently fully supporting and the mean coverage was over 75 percent. Trib. Mineral Fork #1 was impaired in the spring, and had a mean coverage of approximately 42 percent (s.d.= 34).

Based on the difference between controls and test stations, it is possible that Trib. Mineral Fork #1 was impaired by patchy and intermittent fine sediment coverage. Fine sediment intolerant taxa (Zweig and Rabeni 2001) were present in the samples. Taxa such as *Caenis* spp. were among the dominant taxa at both stations. Others, such as *Maccaffertium* (*Stenonema*) spp., *Cricotopus/Orthocladius*, and *Thienemanniella* spp. were present, but in low numbers at both stations. The presence of burrowers (*Ephemera* sp.) illustrate that fine sediment is available. As mentioned earlier, the proportion of sand and silt may have contributed to the conflicting information (Zweig and Rabeni 2001). Fine sediment may have been a contributor to impairment in the spring.

4.2.6 Fine Sediment Character

The fine sediment in the substrate was analyzed for cadmium, lead, and zinc content. These results were compared to threshold limits for individual metals (PEC, MacDonald et al. 2000; PEQ, Besser et al. 2009a), and mixture of metals thresholds (Σ PEQ, and mean PEQ, MacDonald et al. 2009).

Lead and zinc exceeded their respective PECs in the Trib. Mineral Fork #2 fine sediment sample. The lead PEQ was over two and a half, and the zinc exceeded one. Lead and zinc were again above their respective PECs at Trib. Mineral Fork #1. The PEQ for lead was over four, and zinc exceeded a quotient of one. The individual metals exceeded a PEQ threshold, which suggests that there is an increased probability of toxic effects due to heavy metals concentrations (Besser et al. 2009a). It appears that lead or zinc in the fine sediment individually may be contributing to the impairment at Trib. Mineral Fork #1.

The effect from a cumulative mixture of metals (cadmium, lead, and zinc) was calculated for each Trib. Mineral Fork station. The Σ PEQ and mean PEQ for cadmium, lead, and zinc were compared to thresholds identified by MacDonald et al. (2009). The combination of concentrations at Trib. Mineral Fork #2 did not exceed the Σ PEQ threshold, but did exceed the mean PEQ threshold. The Σ PEQ threshold at Trib. Mineral Fork #1 was not reached; however, the mean PEQ was likewise surpassed. Given recommendations by MacDonald et al. (2009) for the mean threshold, fine sediment in Trib. Mineral Fork has a high likelihood of being toxic to benthic macroinvertebrates due to a mixture of heavy metals.

5.0 Summary

Because of the number of streams of interest and in an effort to maintain consistency with the previous study (MDNR 2009a) results are summarized in this section. This summary includes MSCI, BI, Δ MSCI, physicochemical trends, dissolved surface water and pore water metals concentrations, fine sediment percent coverage, and fine sediment character with comparisons of individual and mixture of metals thresholds.

Pond Creek #2

- Fully supporting MSCI score in fall, partially supporting in spring
- Consistently high BI values for both seasons

- Δ MSCI compared to control criteria: no change in either season
- Low conductivity, low organic pollutant indicators in the water quality parameters
- Surface water with elevated barium and nickel in both seasons
- Pore water results not available due to loss of samplers
- Fine sediment coverage was significantly greater than controls (MDNR 2009a)
- Fine sediment character included total recoverable zinc above PEC
- Mixture (Cd, Pb, Zn) metals not greater than threshold

Pond Creek #1

- Fully supporting MSCI scores in both seasons
- Consistently low BI values compared to BIOREF
- Δ MSCI compared to control criteria resulted in slight decrease in fall and no change in spring
- Higher BI values compared to control in both seasons
- Surface water with elevated barium and nickel in both seasons; cadmium elevated in spring
- Pore water with elevated barium
- Fine sediment coverage was significantly greater than controls (MDNR 2009a)
- Fine sediment character included total recoverable zinc above PEC
- Mixture (Cd, Pb, Zn) metals not greater than threshold

Shibboleth Branch #3

- Partially supporting MSCI scores in both seasons
- Consistently low BI values compared to BIOREF
- Δ MSCI compared to control criteria resulted in slight decrease in fall, but no change in spring
- Higher BI values compared to control in both seasons
- Surface water with elevated barium, lead, and nickel in both seasons
- Pore water with elevated barium, cobalt; lead was above chronic WQS
- Fine sediment coverage not significantly greater due to standard deviation (patchy), but SHAPP sediment estimate high and fluctuates (MDNR 2009a)
- Fine sediment character showed total recoverable lead and zinc above PEC
- Mixture (Cd, Pb, Zn) metals greater than Σ PEQ, and mean PEQ threshold

Shibboleth Branch #1

- Fully supporting MSCI score in fall, partially supporting in spring
- Consistently high BI values
- Δ MSCI compared to control criteria resulted in no change in fall, increase from partially to fully supporting in spring
- Low concentrations of organic pollutant indicators in the water quality parameters
- Surface water with elevated barium and nickel in both seasons, zinc in spring
- Pore water with elevated barium and lead
- Fine sediment coverage was significantly greater than controls (MDNR 2009a)
- Fine sediment character showed total recoverable cadmium, lead, and zinc above PEC

- Mixture (Cd, Pb, Zn) metals not greater than Σ PEQ, but greater than mean PEQ threshold

Trib. Mineral Fork #2

- Fully supporting MSCI score in both seasons
- Consistently high BI values
- Δ MSCI compared to control criteria showed no change in either season
- Conductivity high, low organic pollutant indicators in the water quality parameters
- Surface water with elevated barium, nickel, and zinc in both seasons; cadmium and lead were elevated in spring
- Pore water with elevated barium, cobalt, and lead
- Fine sediment coverage was significantly greater than controls
- Fine sediment character showed total recoverable lead and zinc above PEC
- Mixture (Cd, Pb, Zn) metals not greater than Σ PEQ, but greater than mean PEQ threshold

Trib. Mineral Fork #1

- Fully supporting MSCI score in fall, partially supporting in spring
- Consistently low BI values compared to BIORREF
- Δ MSCI compared to control criteria resulted in slight decrease in fall MSCI score, increase from partial to full support in spring
- Higher BI values when compared to control criteria in both seasons
- Conductivity high, low concentrations of organic pollutant indicators
- Surface water with elevated barium and nickel in both seasons, zinc in spring
- Pore water with elevated barium
- Fine sediment coverage was significantly greater than controls
- Fine sediment character showed total recoverable lead above PEC
- Mixture (Cd, Pb, Zn) metals not greater than Σ PEQ, but greater than mean PEQ threshold

6.0 Conclusions

The objectives of this project were met.

Stream habitat was assessed and was found to be similar to controls.

Supportability of the protection of aquatic life beneficial use designation was assessed at these tributaries of Mill Creek and Mineral Fork. Pond Creek #2 was partially supporting in the spring and consistently had high BI values. Shibboleth Branch #3 (upstream) was again consistently impaired. Shibboleth Branch #1 was partially supporting during the spring with high BI values. Trib. Mineral Fork #2 was partially supporting in the spring, with high BI values. Trib. Mineral Fork #1 was impaired during the spring.

Two downstream sites (Shibboleth Branch #1 and Trib. Mineral Fork #1) changed to fully supporting when compared to similar size control streams, which implies a potential stream size influence. Although this change in supportability category suggests that size influenced the scores, the elevated BI values observed when compared to similar size control streams suggested that they were not small control stream quality.

Physicochemical conditions were analyzed. Organic pollutant indicators or nutrients were detected in low levels in surface water samples collected from stations with high BI values. Conductivity was high at Trib. Mineral Fork stations, potentially resulting from mine-related influences.

Surface water contained dissolved metals in low concentrations at several sites. The dissolved metals included barium, nickel, and in one case, consistently higher zinc in the surface water at test sites compared to control streams. None of the concentrations exceeded WQSs.

Although surface water samples did not contain dissolved metals in concentrations above WQSs, pore water samples were sometimes elevated substantially. Barium concentrations were generally high and similar between surface and pore water. However, dissolved cobalt was detected in two streams in pore water but not detected in the surface water. Lead was ten times higher than the chronic WQS in the pore water at one station (Shibboleth Branch #3), yet it was detected in very low concentrations in surface water. The presence of metals in the field blank negated including the results of copper, nickel, and zinc. Evidence suggests that metals in the pore water may have contributed to impairment of Shibboleth Branch #3, and the differences between surface and pore water concentrations suggest that pore water should be sampled using peepers at all mine related streams.

Fine sediment coverage analysis for Mill Creek was conducted in the 2009 MDNR study, which suggested that it may have contributed to impairment of the Pond Creek #2 and Shibboleth Branch #3 macroinvertebrate community. The Mineral Fork tributary was assessed during this study, and fine sediment coverage was significantly greater than the controls at both stations. It may have contributed to the partially supporting status of the downstream station in one season.

Fine sediment character showed total recoverable metals in levels above toxicity thresholds. The sediment at all streams contained at least cadmium, lead, or zinc in varying concentrations above PECs. Low levels in surface water samples suggest that dissolved metals are somewhat labile; however, differences found in one pore water sample (Shibboleth Branch #3) also suggest that metals may be readily available.

Testing of the null hypotheses resulted in the following:

- 1) Stream habitat quality was similar among tributaries and controls.

- 2) Biological metrics revealed that several tributaries were partially supporting of the AQL beneficial use designation.
- 3) Physicochemical results identified potential organic influences; however, none exceeded WQSs.
- 4) Surface water dissolved metals were elevated compared to controls, and did not exceed WQSs.
- 5) Pore water metals were elevated compared to controls, and one station exceeded WQS for lead by a factor of ten.
- 6) Fine sediment coverage was greater than the controls at all but one station, which had a high standard deviation of data due to patchy and intermittent distribution.
- 7) Fine sediment character revealed that cadmium, lead, or zinc were present in fine sediment above threshold levels for individual and mixtures of total recoverable metals

7.0 Recommendations

- 1) Sample pore water using peepers (Brumbaugh et al. 2002, 2007) as standard procedure in future mine related stream studies.
- 2) Sample pore water using peepers (Brumbaugh et al. 2002, 2007) at mine related streams where biological assessments and stream studies have already been completed.

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Appendix A

Macroinvertebrate Database Bench Sheet Report for Mill Creek Tributaries and Trib.
Mineral Fork Stations, Fall 2010 – Spring 2011

Order: Pond Creek, Shibboleth Branch, Trib. Mineral Fork

Aquid Invertebrate Database Bench Sheet Report

Pond Cr [1004018], Station #2, Sample Date: 9/22/2010 8:45:00 AM

CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	21	4	4
AMPHIPODA			
Gammarus			19
COLEOPTERA			
Dubiraphia		2	47
Ectopria nervosa	1	-99	
Helichus lithophilus			1
Microcylloepus pusillus	1		
Optioservus sandersoni	3		1
Psephenus herricki		1	
Stenelmis	5		
DECAPODA			
Orconectes medius	2		
Orconectes punctimanus		-99	1
DIPTERA			
Ablabesmyia	1	3	
Antocha			1
Ceratopogoninae	3	1	
Chrysops		-99	
Cladotanytarsus	1	3	
Clinocera	1		
Corynoneura	8	4	1
Cricotopus bicinctus	2		1
Cricotopus/Orthocladius	24	2	
Dixa	1		9
Djalmabatista			1
Forcipomyiinae	1	1	
Hemerodromia	21	3	15
Microtendipes		4	1
Natarsia	1		
Paracricotopus	1		
Parakiefferiella	2	2	
Parametriocnemus	7		1
Paraphaenocladius			1
Paratanytarsus	1	2	18
Phaenopsectra		1	
Polypedilum aviceps	2		
Polypedilum illinoense grp	8		3
Procladius			1
Rheocricotopus	3		

Aquid Invertebrate Database Bench Sheet Report

Pond Cr [1004018], Station #2, Sample Date: 9/22/2010 8:45:00 AM

CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence

ORDER: TAXA	CS	NF	RM
Rheotanytarsus	20		3
Simulium	9		7
Stempellina		1	
Stempellinella	21	18	2
Stenochironomus	1	1	2
Tabanus	1		
Tanytarsus	20	33	12
Thienemanniella	8	2	3
Thienemannimyia grp.	8	2	4
Tipula	4		
Tribelos	6	2	
Tvetenia bavarica grp	3		1
undescribed Empididae			3
EPHEMEROPTERA			
Baetis	4		
Caenis anceps	1		
Caenis latipennis	336	168	129
Centroptilum		2	
Diphetor			2
Ephemera simulans	1	2	
Eurylophella	3	2	4
Hexagenia limbata		3	
Isonychia bicolor	22		
Leptophlebiidae	1		3
Maccaffertium pulchellum	50		1
Stenacron	7	3	
Stenonema femoratum	2	2	
HEMIPTERA			
Microvelia	5		1
Rhagovelia			1
ISOPODA			
Caecidotea	2		24
LEPIDOPTERA			
Petrophila	2		
LIMNOPHILA			
Lymnaeidae	1		1
MEGALOPTERA			
Corydalus	3		
Nigronia serricornis	1		
ODONATA			
Argia	10	6	15

Aquid Invertebrate Database Bench Sheet Report

Pond Cr [1004018], Station #2, Sample Date: 9/22/2010 8:45:00 AM

CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence

ORDER: TAXA	CS	NF	RM
Boyeria			1
Calopteryx			2
Enallagma			1
Gomphidae	1		3
Hagenius brevistylus	3	5	2
Hetaerina			2
Stylogomphus albistylus	-99	-99	
TRICHOPTERA			
Cheumatopsyche	43		6
Chimarra	7		
Helicopsyche	5		
Hydropsyche			1
Limnephilidae		2	3
Nectopsyche			2
Oecetis	1		15
Polycentropodidae		1	
Polycentropus	3		
Ptilostomis			2
Pycnopsyche			2
Triaenodes			10
TRICLADIDA			
Planariidae			1
TUBIFICIDA			
Aulodrilus		3	1
Limnodrilus hoffmeisteri		1	
Tubificidae		7	3
VENEROIDA			
Pisidiidae	2	3	22

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [1004019], Station #1, Sample Date: 9/22/2010 10:35:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	14	7	21
AMPHIPODA			
Hyaella azteca			3
COLEOPTERA			
Dubiraphia		15	91
Ectopria nervosa	2	2	
Helichus basalis			1
Heterosternuta			1
Microcylloepus pusillus		2	12
Optioservus sandersoni	17	1	2
Paracymus			1
Psephenus herricki	18	2	
Stenelmis	2		19
DECAPODA			
Orconectes medius	3		
DIPTERA			
Ablabesmyia		2	
Anopheles			2
Ceratopogoninae		9	
Chrysops		1	
Cladotanytarsus		1	
Corynoneura			2
Cricotopus bicinctus			2
Cricotopus/Orthocladius	7		6
Cryptochironomus		2	
Cryptotendipes		2	
Dixella			1
Ephydriidae		1	
Forcipomyiinae	1		1
Hemerodromia	1	2	
Labrundinia			1
Nanocladius			1
Parakiefferiella		1	
Paralauterborniella		4	
Paratanytarsus		2	2
Paratendipes		1	
Polypedilum aviceps	1		
Polypedilum convictum	3		
Polypedilum halterale grp			6

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [1004019], Station #1, Sample Date: 9/22/2010 10:35:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Procladius		2	
Prosimulium	2		
Rheotanytarsus			4
Silvius	-99		
Stempellina		1	
Stempellinella	1	39	8
Stenochironomus			1
Stictochironomus		1	
Tabanus	-99		
Tanytarsus		23	4
Thienemanniella	1	1	
Thienemannimyia grp.	3	3	4
Tribelos		25	
Zavrelimyia		1	
EPHEMEROPTERA			
Acentrella	2		
Baetis	4		
Baetisca lacustris	1	1	
Caenis anceps	25	2	
Caenis latipennis	59	113	18
Ephemera simulans	1	2	
Eurylophella	162	15	9
Isonychia	37		
Leptophlebiidae	3	4	
Maccaffertium mediopunctatum	6		
Maccaffertium pulchellum	82	3	
Procloeon		5	2
Stenacron	16	5	
Stenonema femoratum	1	8	
Tricorythodes	13		
LEPIDOPTERA			
Crambidae	1		
LIMNOPHILA			
Ancylidae		14	1
Lymnaeidae			1
Menetus			3
LUMBRICINA			
Lumbricina	17		
MEGALOPTERA			
Corydalus	1		
Nigronia serricornis	4		-99

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [1004019], Station #1, Sample Date: 9/22/2010 10:35:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Sialis		1	
MESOGASTROPODA			
Elimia	3		1
ODONATA			
Argia	11	7	10
Enallagma			5
Gomphidae	109	2	1
Hagenius brevistylus			1
Stylogomphus albistylus		1	
TRICHOPTERA			
Cernotina	1		
Cheumatopsyche	12		
Chimarra	1		
Helicopsyche		1	
Hydropsyche	4		
Leptoceridae		4	
Oecetis			9
Polycentropus	3		
Triaenodes			4
TUBIFICIDA			
Enchytraeidae			1
Tubificidae		21	1
VENEROIDA			
Pisidiidae	1	2	7

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004020], Station #3, Sample Date: 9/22/2010 1:00:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	5	10	3
COLEOPTERA			
Dubiraphia		4	16
Ectopria nervosa		2	
Macronychus glabratus		3	6
Microcylloepus pusillus	1	1	
Optioservus sandersoni	10		
Stenelmis	2		
DECAPODA			
Orconectes medius	-99	-99	
Orconectes punctimanus		1	
DIPTERA			
Ablabesmyia		12	5
Ceratopogoninae		3	
Chironomidae		2	
Chrysops		-99	
Cladotanytarsus	1	2	
Corynoneura	2		
Cricotopus bicinctus			1
Cricotopus/Orthocladius		1	
Cryptochironomus		1	
Diptera		1	1
Dixa	2		
Forcipomyiinae	1		
Hemerodromia	2	4	16
Labrundinia	1		1
Microtendipes		4	2
Myxosargus	1	1	1
Nanocladius	1	1	
Parakiefferiella	1	24	1
Parametriocnemus			1
Paratanytarsus	2	6	75
Paratendipes		1	
Phaenopsectra		3	
Polypedilum aviceps	5		
Polypedilum fallax grp	1	1	
Polypedilum illinoense grp	14	12	21
Rheotanytarsus	6	2	10
Simulium	11		1

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004020], Station #3, Sample Date: 9/22/2010 1:00:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Stempellinella	3	3	
Stenochironomus	1	1	1
Tanytarsus	45	213	84
Thienemanniella	5	6	1
Thienemannimyia grp.	2	2	5
Tipula	6		
Tribelos			2
EPHEMEROPTERA			
Baetis	30		
Caenis latipennis	2	4	
Eurylophella		1	2
Isonychia bicolor	14		
Maccaffertium pulchellum	284	3	1
Stenacron	12	1	
HEMIPTERA			
Microvelia	1		
Rhagovelia	3		
ISOPODA			
Caecidotea	3	1	1
LIMNOPHILA			
Ancylidae		2	
Lymnaeidae		1	1
MEGALOPTERA			
Nigronia serricornis	5	-99	-99
Sialis		-99	
ODONATA			
Basiaeschna janata			-99
Boyeria	-99		1
Calopteryx		3	17
Gomphidae	1		
Hagenius brevistylus	2	13	3
Hetaerina			4
TRICHOPTERA			
Cheumatopsyche	36		
Chimarra	20		
Oecetis			9
Oxyethira			2
Polycentropus	12	1	5
Ptilostomis			1
Triaenodes			43
TUBIFICIDA			

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004020], Station #3, Sample Date: 9/22/2010 1:00:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Branchiura sowerbyi		1	
Tubificidae		1	2
VENEROIDA			
Pisidiidae		1	

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004017], Station #1, Sample Date: 9/21/2010 2:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	7	6	19
AMPHIPODA			
Hyaella azteca			5
BRANCHIOBDELLIDA			
Branchiobdellida	4		
COLEOPTERA			
Coptotomus			-99
Dubiraphia		5	55
Macronychus glabratus		1	5
Optioservus sandersoni	6	1	
Psephenus herricki		1	
Stenelmis	62	2	3
DECAPODA			
Cambarus maculatus	1		
Orconectes harrisonii	-99		
Orconectes medius	-99		
Orconectes punctimanus	1		
DIPTERA			
Ablabesmyia		6	2
Ceratopogoninae		7	1
Chironomidae		1	1
Chironomus		3	
Cladopelma		7	1
Cladotanytarsus		22	5
Corynoneura	1		
Cricotopus/Orthocladius	6		1
Cryptochironomus		3	
Cryptotendipes		6	3
Culicidae			1
Dicrotendipes	2	32	
Diptera	1		
Empididae			1
Hemerodromia	4		
Labrundinia			3
Microtendipes			3
Nanocladius		1	
Nilotanypus	4		
Paracladopelma		2	
Parakiefferiella		3	1

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004017], Station #1, Sample Date: 9/21/2010 2:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Parametriocnemus	3		
Paratanytarsus	1	1	1
Polypedilum aviceps	12		
Polypedilum convictum	2		
Polypedilum illinoense grp	4	4	1
Polypedilum scalaenum grp		1	
Procladius		2	2
Rheotanytarsus	16		
Simulium	6		
Stempellina		4	
Stempellinella	17		12
Stenochironomus			3
Tanytarsus	36	77	61
Thienemanniella	9	1	
Thienemannimyia grp.	6		1
Tribelos		10	3
Tvetenia	1		
EPHEMEROPTERA			
Baetis	15		
Caenis anceps	64	2	1
Caenis latipennis	150	22	46
Centroptilum		5	6
Ephemera simulans	4	-99	
Ephemerella needhami			1
Eurylophella	12	1	
Eurylophella bicolor			1
Heptageniidae	23		1
Hexagenia		3	3
Isonychia bicolor	38		
Leptophlebiidae	2	1	4
Maccaffertium mediopunctatum	18		
Maccaffertium pulchellum	29		
Stenacron	6		
Tricorythodes	18		
HEMIPTERA			
Rhagovelia	1		
ISOPODA			
Caecidotea	34	1	
LIMNOPHILA			
Ancylidae			1
LUMBRICINA			

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [1004017], Station #1, Sample Date: 9/21/2010 2:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Lumbricina		-99	
MEGALOPTERA			
Corydalus	2		
Nigronia serricornis	4		
Sialis		-99	
MESOGASTROPODA			
Elimia	2		
ODONATA			
Argia	9	2	5
Didymops		1	1
Enallagma			16
Gomphidae	1		
Macromia			-99
Stylogomphus albistylus			1
TRICHOPTERA			
Cheumatopsyche	16		
Chimarra	6		
Helicopsyche	2		
Mystacides		9	
Polycentropus	6		
Ptilostomis			1
Triaenodes	1		11
TRICLADIDA			
Planariidae	2	1	
TUBIFICIDA			
Tubificidae	3		6
VENEROIDA			
Corbicula	3	4	

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004015], Station #2, Sample Date: 9/21/2010 10:38:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	5	1	3
AMPHIPODA			
Gammarus	-99		6
Hyaella azteca			1
BRANCHIOBDELLIDA			
Branchiobdellida			1
COLEOPTERA			
Dubiraphia	1	13	19
Ectopria nervosa		3	
Helichus basalis			1
Macronychus glabratus			1
Microcylloepus pusillus	27	1	12
Optioservus sandersoni	37		
Psephenus herricki	22	4	
Stenelmis	69		14
DECAPODA			
Orconectes harrisonii		-99	
Orconectes medius	1		-99
Orconectes punctimanus	-99		
Orconectes virilis		1	
DIPTERA			
Ablabesmyia		6	1
Anopheles			3
Ceratopogoninae		8	1
Chironomidae	1	1	
Chironomus	1		
Cladopelma		1	
Clinotanypus			2
Corynoneura	2		2
Cricotopus/Orthocladius	2		1
Cryptochironomus		2	
Cryptotendipes		16	
Dicrotendipes		1	
Dixella			1
Hemerodromia	9		
Labrundinia	1	2	7
Larsia		1	
Microtendipes	6	2	4
Myxosargus			2

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004015], Station #2, Sample Date: 9/21/2010 10:38:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Natarsia		3	2
Nilotanypus	4		
Parakiefferiella		1	
Paralauterborniella		8	
Parametrioconemus	5		
Paratanytarsus			15
Polypedilum aviceps	13		2
Polypedilum halterale grp		11	
Polypedilum illinoense grp	2		3
Polypedilum scalaenum grp	2		
Procladius		2	
Rheocricotopus	13		
Rheotanytarsus	20		5
Simulium	9		
Stempellinella	21	7	10
Stenochironomus			2
Stictochironomus		3	
Stratiomyidae		1	
Tabanus	1		
Tanytarsus	12	31	14
Thienemanniella	10		2
Thienemannimyia grp.	14	1	16
Tipula			-99
Tribelos	1	5	
Tvetenia bavarica grp	1		
EPHEMEROPTERA			
Acerpenna	1		
Baetis	11		
Caenis anceps	3	35	8
Caenis latipennis	29	287	35
Centroptilum	1		
Ephemera simulans		1	
Isonychia bicolor	11		
Leptophlebiidae	3	1	
Maccaffertium mediopunctatum	2		
Maccaffertium pulchellum	52		1
Stenacron	7		
Stenonema femoratum		2	
Tricorythodes	3		
HEMIPTERA			
Ranatra fusca			1

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004015], Station #2, Sample Date: 9/21/2010 10:38:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Rhagovelia	1		
ISOPODA			
Caecidotea	21		30
LIMNOPHILA			
Ancylidae	2		4
Lymnaeidae			1
Menetus			2
LUMBRICINA			
Lumbricina	4		1
MEGALOPTERA			
Corydalus	1		
Nigronia serricornis	2		-99
Sialis		-99	
MESOGASTROPODA			
Elimia	12	3	7
ODONATA			
Argia			14
Calopteryx	1		3
Gomphidae	31		
PLECOPTERA			
Acroneuria	1		
Neoperla	2		
TRICHOPTERA			
Cheumatopsyche	41		
Chimarra	53		
Helicopsyche	1		
Hydroptilidae			1
Oecetis	3	1	4
Polycentropus	1		
Ptilostomis			1
Triaenodes			2
TRICLADIDA			
Planariidae	9	1	6
TUBIFICIDA			
Branchiura sowerbyi		5	2
Tubificidae	1	32	7
VENEROIDA			
Pisidiidae	3	5	

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004016], Station #1, Sample Date: 9/21/2010 11:45:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	2		1
AMPHIPODA			
Hyaella azteca			1
BRANCHIOBDELLIDA			
Branchiobdellida			1
COLEOPTERA			
Dubiraphia		16	33
Ectopria nervosa		4	
Helichus basalis			1
Macronychus glabratus			3
Microcylloepus pusillus	16	1	40
Optioservus sandersoni	11	1	1
Psephenus herricki	28	6	
Scirtidae			1
Stenelmis	82	20	7
DECAPODA			
Orconectes medius	1		-99
DIPTERA			
Ablabesmyia	1	6	4
Anopheles			1
Brillia	1		
Ceratopogoninae		2	1
Chironomidae	2	1	1
Cladotanytarsus	1	1	4
Clinotanypus			1
Cricotopus/Orthocladius	2		1
Cryptochironomus		1	
Cryptotendipes		2	
Dicrotendipes			1
Hemerodromia	13	1	6
Labrundinia	1	1	5
Microtendipes	1	3	2
Natarsia		2	
Parametriocnemus	20		
Paratanytarsus		2	25
Phaenopsectra		2	
Polypedilum aviceps	76		6
Polypedilum halterale grp			2
Polypedilum illinoense grp	5	1	2

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004016], Station #1, Sample Date: 9/21/2010 11:45:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Polypedilum scalaenum grp	1		
Procladius			1
Rheocricotopus	24		3
Rheotanytarsus	21	1	
Simulium	7		1
Stempellinella	20	16	16
Stenochironomus	1	2	2
Stictochironomus		1	
Tabanus	-99		
Tanytarsus	20	26	43
Thienemanniella	2		10
Thienemannimyia grp.	16	4	4
Tribelos		5	
undescribed Empididae	5		
Zavrelimyia	1		
EPHEMEROPTERA			
Acerpenna	2		
Baetis	29		1
Caenis anceps	28	4	
Caenis latipennis	6	95	36
Ephemera simulans		2	1
Isonychia bicolor	26		
Leptophlebiidae	4	18	9
Maccaffertium mediopunctatum	1		
Maccaffertium pulchellum	244		
Stenacron	4	8	1
Stenonema femoratum		16	4
HEMIPTERA			
Microvelia	3		2
Rhagovelia	3		
Trepobates			1
ISOPODA			
Caecidotea	17	5	11
LIMNOPHILA			
Ancylidae	3	19	13
Menetus	2	2	
Physella			-99
LUMBRICINA			
Lumbricina	2		1
MEGALOPTERA			
Corydalus	1		

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [1004016], Station #1, Sample Date: 9/21/2010 11:45:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Nigronia serricornis	12	-99	
Sialis		1	
MESOGASTROPODA			
Elimia	-99		
ODONATA			
Argia	4	8	9
Calopteryx			2
Gomphidae	11	1	1
Libellula			1
Stylogomphus albistylus		-99	
PLECOPTERA			
Acroneuria	-99	2	
Neoperla	10		
Perlesta	25		
Zealeuctra	1		1
TRICHOPTERA			
Cheumatopsyche	26		1
Chimarra	74		1
Hydropsyche	1		
Oecetis		1	
Orthotrichia			1
Polycentropus	1	1	
Pycnopsyche			-99
Triaenodes			3
TUBIFICIDA			
Branchiura sowerbyi		3	1
Tubificidae		7	10
VENEROIDA			
Pisidiidae		1	2

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110323], Station #2, Sample Date: 3/23/2011 10:05:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	11	17	2
AMPHIPODA			
Gammarus	5	5	15
BRANCHIOBDELLIDA			
Branchiobdellida	2	1	
COLEOPTERA			
Dubiraphia	1	7	6
Ectopria nervosa	1	1	
Microcylloepus pusillus			1
Stenelmis	1		1
DECAPODA			
Orconectes medius	3	1	-99
Orconectes punctimanus		-99	
DIPTERA			
Ablabesmyia		5	1
Antocha	1		
Ceratopogoninae		15	7
Chaetocladius	1		
Chelifera	2		
Chironomidae	3		
Cladotanytarsus		3	1
Corynoneura		2	1
Cricotopus bicinctus	1		2
Cricotopus/Orthocladius	14	5	3
Cryptochironomus		6	
Dasyheleinae		1	
Dicrotendipes		1	
Diptera	4	5	1
Djalmabatista		1	
Epoicocladius		1	
Eukiefferiella	8		2
Gonomyia		1	
Hemerodromia	53	4	13
Heterotrissocladius		1	
Limnophyes		1	
Micropsectra			4
Microtendipes		1	2
Parakiefferiella		11	
Paralauterborniella	1	3	

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110323], Station #2, Sample Date: 3/23/2011 10:05:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Parametrioctenemus	5		1
Paratanytarsus			9
Phaenopsectra		4	
Pilaria		2	
Polypedilum aviceps	12		
Polypedilum scalaenum grp		1	
Procladius		2	
Prosimulium	24		
Psectrocladius		1	
Pseudolimnophila		1	
Rheocricotopus	1		1
Rheotanytarsus	4	1	1
Simulium	47		7
Stegopterna	1		
Stempellina	23	6	7
Tanytarsus	12	29	10
Thienemanniella	1	1	5
Thienemannimyia grp.	4	7	6
Tipula	2	-99	1
Tvetenia	2		3
undescribed Empididae			1
Zavrelimyia		2	
EPHEMEROPTERA			
Caenis latipennis	247	173	131
Ephemera simulans		1	
Eurylophella	2		1
Hexagenia limbata		1	
Isonychia bicolor	13		
Leptophlebia			1
Maccaffertium pulchellum	63		
Stenacron	8	4	
Stenonema femoratum	1	2	
HEMIPTERA			
Microvelia		1	1
ISOPODA			
Caecidotea	4	2	6
LEPIDOPTERA			
Petrophila	1		
LIMNOPHILA			
Lymnaeidae	1		
Menetus	1		

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110323], Station #2, Sample Date: 3/23/2011 10:05:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
MEGALOPTERA			
Corydalis	1		
Nigronia serricornis	1		2
ODONATA			
Argia	3		1
Gomphidae		2	
Gomphus		-99	
Hagenius brevistylus		1	1
Stylogomphus albistylus		-99	
PLECOPTERA			
Amphinemura	3		3
Clioperla clio			1
Leuctridae	26	1	
TRICHOPTERA			
Cheumatopsyche	11		7
Chimarra	6		
Hydroptila			3
Oecetis		2	1
Polycentropus			2
Ptilostomis			1
Pycnopsyche		2	-99
Triaenodes			6
TRICLADIDA			
Planariidae	1		
TUBIFICIDA			
Enchytraeidae		1	1
Limnodrilus hoffmeisteri		6	
Tubificidae		28	
VENEROIDA			
Pisidiidae	2	2	3

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110324], Station #1, Sample Date: 3/23/2011 11:40:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	43	23	
BRANCHIOBDELLIDA			
Branchiobdellida	1		
COLEOPTERA			
Dubiraphia		14	32
Ectopria nervosa		3	-99
Heterosternuta		1	
Optioservus sandersoni	10	2	2
Psephenus herricki	1		
Stenelmis		1	
DECAPODA			
Cambarus maculatus			1
Orconectes medius	2		1
DIPTERA			
Ablabesmyia		4	2
Ceratopogoninae		7	
Chironomidae	1		1
Cladotanytarsus	1	1	
Clinocera	33	6	
Corynoneura	12	2	14
Cricotopus bicinctus	1		
Cricotopus/Orthocladius	21		11
Cryptochironomus	1	1	1
Dasyheleinae			1
Dicrotendipes		1	
Dixella			1
Eukiefferiella	16	2	3
Hemerodromia	3		2
Labrundinia			10
Limnophyes	1		
Micropsectra	5	2	3
Microtendipes		1	
Nanocladius	1		
Orthocladius (Euorthocladius)	7		
Parakiefferiella	6	17	9
Paralauterborniella		2	
Parametriocnemus	4		
Paratanytarsus			2
Phaenopsectra	1		

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110324], Station #1, Sample Date: 3/23/2011 11:40:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Polypedilum aviceps	11		
Polypedilum illinoense grp			1
Polypedilum scalaenum grp	1	2	
Prosimulium	10		
Pseudorthocladius			1
Rheocricotopus	8		
Rheotanytarsus	13		3
Simulium	23	1	
Stempellina		2	
Stempellinella	31	8	10
Tabanus	1		
Tanytarsus	44	13	14
Thienemanniella	13	1	9
Thienemannimyia grp.	14	11	14
Tipula	1		
Tribelos		2	
Tvetenia	3		
Zavrelimyia		1	
EPHEMEROPTERA			
Acerpenna			1
Caenis latipennis	83	111	107
Ephemera simulans		2	
Eurylophella bicolor	63	39	29
Eurylophella enoensis			2
Heptageniidae	9		
Isonychia bicolor	9		
Leptophlebiidae	1		1
Maccaffertium mediopunctatum	9		
Maccaffertium pulchellum	34	3	
Procloeon			4
Stenacron	26	3	1
Stenonema femoratum	4		1
HEMIPTERA			
Microvelia			2
LIMNOPHILA			
Lymnaeidae		2	1
LUMBRICINA			
Lumbricina	4		
MEGALOPTERA			
Nigronia serricornis	3		
MESOGASTROPODA			

Aquid Invertebrate Database Bench Sheet Report**Pond Cr [110324], Station #1, Sample Date: 3/23/2011 11:40:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Elimia	1		1
ODONATA			
Argia		1	
Enallagma			1
Gomphidae	11	7	3
Hagenius brevistylus			-99
PLECOPTERA			
Amphinemura	7		1
Leuctridae	43	8	
Perlesta	6		
Prostoia	3		
TRICHOPTERA			
Cheumatopsyche	7		2
Chimarra	8		
Helicopsyche	3	1	1
Micrasema			3
Mystacides		3	2
Oecetis		1	2
Polycentropus	4	2	
Pycnopsyche	1		3
Rhyacophila	11		
Triaenodes			1
TRICLADIDA			
Planariidae	8		1
TUBIFICIDA			
Enchytraeidae		1	1
Tubificidae			1
VENEROIDA			
Pisidiidae		2	

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110328], Station #3, Sample Date: 3/24/2011 1:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	8	25	2
BRANCHIOBDELLIDA			
Branchiobdellida			1
COLEOPTERA			
Dubiraphia			3
Macronychus glabratus			1
Optioservus sandersoni	5	3	
Scirtidae	1	1	
DECAPODA			
Orconectes medius	-99	1	-99
Orconectes punctimanus			1
DIPTERA			
Ablabesmyia		4	1
Antocha	1		
Brillia	2		
Ceratopogoninae		17	3
Chironomidae	2	1	
Clinocera	7	2	
Corynoneura	2	3	2
Cricotopus bicinctus	6		13
Cricotopus/Orthocladius	33	5	13
Cryptochironomus	2	1	
Diptera		1	
Dixa		1	
Hemerodromia	13	5	11
Limnophyes	1		
Microtendipes			1
Myxosargus	1		
Nanocladius		1	1
Orthocladius (Euorthocladius)	1		
Parakiefferiella	19	34	6
Parametriocnemus	6		1
Paraphaenocladius	2	1	
Paratanytarsus			10
Polypedilum aviceps	57	1	4
Polypedilum illinoense grp	1	2	1
Procladius			1
Prosimulium	5		
Rheocricotopus	3	1	2

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110328], Station #3, Sample Date: 3/24/2011 1:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Rheotanytarsus	5		2
Simulium	210	5	6
Stegopterna	1		
Stempellinella	11	2	6
Tanytarsus	63	84	78
Thienemanniella	15	6	29
Thienemannimyia grp.	6	3	3
Tipula	3		
Tvetenia	2	1	
EPHEMEROPTERA			
Caenis latipennis	3	3	5
Eurylophella enoensis		1	2
Isonychia bicolor	8		
Maccaffertium pulchellum	174	19	14
Stenacron	8	18	
ISOPODA			
Caecidotea	6	2	6
LIMNOPHILA			
Lymnaeidae			1
Menetus			3
MEGALOPTERA			
Nigronia serricornis	3		-99
ODONATA			
Boyeria			1
Calopteryx			5
Enallagma			1
Hagenius brevistylus		2	1
Stylogomphus albistylus		-99	
PLECOPTERA			
Amphinemura	19	1	2
Capniidae		1	
Clioperla clio	-99		
Leuctridae	6	3	2
Perlidae			1
TRICHOPTERA			
Cheumatopsyche	28		1
Chimarra	20		
Hydroptila	1		1
Neureclipsis	5	1	5
Oecetis			1
Oxyethira		2	16

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110328], Station #3, Sample Date: 3/24/2011 1:20:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Pycnopsyche		-99	
Rhyacophila	9		
Triaenodes			16
TUBIFICIDA			
Branchiura sowerbyi		1	
Tubificidae		4	
VENEROIDA			
Pisidiidae	1	2	1

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110325], Station #1, Sample Date: 3/23/2011 1:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	33	14	2
AMPHIPODA			
Hyaella azteca			3
COLEOPTERA			
Dubiraphia	2	4	31
Macronychus glabratus			1
Microcylloepus pusillus	2		1
Optioservus sandersoni	7		
Scirtidae			1
Stenelmis	30		2
DECAPODA			
Orconectes hylas	1		
Orconectes luteus	-99		
DIPTERA			
Ablabesmyia		6	22
Atherix	1		
Brillia	1		
Ceratopogoninae		4	
Chironomidae	1	3	1
Chironomus			1
Cladotanytarsus		7	
Clinocera	1		
Clinotanypus		1	
Corynoneura		1	1
Cricotopus bicinctus	4		5
Cricotopus/Orthocladius	41	3	3
Cryptochironomus	1	7	
Cryptotendipes		1	
Dicrotendipes		1	3
Epoicocladius		2	
Eukiefferiella	8		
Forcipomyiinae			1
Hemerodromia	12	2	7
Labrundinia			3
Microtendipes	5	4	4
Myxosargus	1		
Nanocladius	1	2	
Parachironomus		1	
Parakiefferiella		11	15

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110325], Station #1, Sample Date: 3/23/2011 1:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Paralauterborniella		3	
Parametriocnemus	5		
Paraphaenocladus			3
Paratanytarsus	1	3	42
Paratendipes		1	
Phaenopsectra		5	1
Polypedilum aviceps	4		
Polypedilum convictum	4	1	
Polypedilum illinoense grp	1	1	1
Polypedilum scalaenum grp	1	6	1
Procladius		1	2
Rheocricotopus	5		
Rheotanytarsus	12	1	
Simulium	5		
Stempellina		6	
Stempellinella	23	13	4
Stictochironomus	1	1	
Tabanus	-99		
Tanypus		1	
Tanytarsus	53	120	56
Thienemanniella			2
Thienemannimyia grp.	13	3	3
Tipula	2		
Tvetenia bavarica grp	2		
EPHEMEROPTERA			
Baetisca lacustris	1		
Caenis latipennis	105	83	71
Centroptilum		-99	2
Ephemera simulans	-99	-99	
Eurylophella enoensis	7		
Hexagenia limbata		-99	1
Isonychia bicolor	52		
Leptophlebiidae	13	1	
Maccaffertium mediopunctatum	10		
Maccaffertium pulchellum	29	1	
Stenacron	18	2	
Stenonema femoratum		2	
Tricorythodes	3		
GORDIOIDEA			
Chordodidae	-99		
ISOPODA			

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110325], Station #1, Sample Date: 3/23/2011 1:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Caecidotea	15	1	1
Caecidotea (Blind & Unpigmented)		1	
LIMNOPHILA			
Ancylidae	2	1	2
LUMBRICINA			
Lumbricina			1
MEGALOPTERA			
Corydalus	1		
Nigronia serricornis	2		
ODONATA			
Argia	1	1	2
Calopteryx			-99
Enallagma			1
Gomphidae	1		
Gomphus			1
Macromia			-99
Stylogomphus albistylus	-99		
PLECOPTERA			
Acroneuria	-99		
Amphinemura	1		
Leuctridae	5		
Perlidae	1		
TRICHOPTERA			
Cheumatopsyche	10		
Chimarra	7		
Helicopsyche	1		
Hydroptila	1		
Mystacides		2	1
Neophylax	1		
Neureclipsis	1		1
Oecetis	1	2	1
Pycnopsyche		-99	-99
Rhyacophila	2		
Setodes		3	2
Triaenodes			1
TRICLADIDA			
Planariidae	1	1	3
TUBIFICIDA			
Aulodrilus		2	2
Enchytraeidae		1	

Aquid Invertebrate Database Bench Sheet Report**Shibboleth Br [110325], Station #1, Sample Date: 3/23/2011 1:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Limnodrilus hoffmeisteri			1
Spirosperma			1
Tubificidae	1	2	2
VENEROIDA			
Corbicula	4	-99	
Pisidiidae	1	3	

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110327], Station #2, Sample Date: 3/24/2011 12:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	23	3	3
AMPHIPODA			
Gammarus	20	3	5
BRANCHIOBDELLIDA			
Branchiobdellida			7
COLEOPTERA			
Dubiraphia	3	5	18
Ectopria nervosa	3	1	2
Lutrochus		1	
Macronychus glabratus	3		
Microcylloepus pusillus	5		
Optioservus sandersoni	16		
Stenelmis	49	6	9
DECAPODA			
Orconectes luteus		-99	
Orconectes medius	2	-99	-99
DIPTERA			
Ablabesmyia		5	
Ceratopogoninae	11	12	3
Chironomidae	2		1
Cladotanytarsus		1	1
Clinocera	3		1
Corynoneura	7	1	8
Cricotopus bicinctus	1		5
Cricotopus/Orthocladius	7		29
Cryptochironomus	3	5	
Cryptotendipes		1	1
Dicrotendipes		1	
Diptera			1
Epoicocladius	1		
Eukiefferiella	19		2
Hemerodromia	20		1
Labrundinia			7
Microtendipes		4	
Nanocladius	1		
Nemotelus			1
Nilothauma		1	
Paracladopelma		1	
Paralauterborniella		11	

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110327], Station #2, Sample Date: 3/24/2011 12:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Parametrioctenemus	12		
Paratanytarsus			5
Phaenopsectra		1	
Polypedilum aviceps	1		4
Polypedilum convictum	8		2
Polypedilum halterale grp	1	1	
Polypedilum illinoense grp	1		8
Polypedilum scalaenum grp	6	5	
Prosimulium	26		
Pseudorthocladius			3
Rheocricotopus	20		3
Rheotanytarsus	10		20
Simulium	8		5
Stempellina		1	
Stempellinella	96	112	21
Stictochironomus		4	
Tabanus	-99		
Tanytarsus	19	33	13
Thienemanniella	7	2	15
Thienemannimyia grp.	26	16	17
Tipula	1		
Tribelos		3	
Tvetenia	3		
Zavreliomyia		5	
EPHEMEROPTERA			
Acerpenna	1		1
Caenis latipennis	152	83	58
Dipheter	2		
Ephemera simulans	-99	-99	
Eurylophella enoensis	2	1	6
Maccaffertium mediopunctatum	3		
Maccaffertium pulchellum	18		
Stenacron	6	5	
Stenonema femoratum		-99	
ISOPODA			
Caecidotea	10		4
LIMNOPHILA			
Ancylidae	1		
MEGALOPTERA			
Nigronia serricornis	-99		
MESOGASTROPODA			

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110327], Station #2, Sample Date: 3/24/2011 12:30:00 PM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Elimia	37	1	9
ODONATA			
Argia			1
Calopteryx			-99
Enallagma			2
Gomphidae	3		
Libellulidae			2
PLECOPTERA			
Acroneuria	-99	-99	
Amphinemura	2		
Leuctridae	6		
Perlidae	10		
TRICHOPTERA			
Cheumatopsyche	5		1
Chimarra	17		
Helicopsyche	11		
Hydroptila	3		
Micrasema	2		
Neophylax	1		-99
Neureclipsis	1		
Oecetis	2		
Pycnopsyche	3	1	3
Rhyacophila	5		
Triaenodes			2
TRICLADIDA			
Planariidae	6		
TUBIFICIDA			
Enchytraeidae	1	2	
Tubificidae	3	13	1
VENEROIDA			
Pisidiidae	10	3	1

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110326], Station #1, Sample Date: 3/24/2011 10:30:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
"HYDRACARINA"			
Acarina	11	7	5
AMPHIPODA			
Gammarus		1	
Hyaella azteca			1
BRANCHIOBDELLIDA			
Branchiobdellida		3	
COLEOPTERA			
Dubiraphia	2	3	7
Ectopria nervosa	1	1	
Heterosternuta		1	
Microcylloepus pusillus	40	2	3
Psephenus herricki	4	1	
Stenelmis	49	17	1
DECAPODA			
Orconectes medius	-99	-99	1
DIPTERA			
Ablabesmyia			1
Ceratopogoninae	1	1	1
Chironomidae	2		2
Chrysops		-99	
Cladotanytarsus		1	
Clinocera	9	1	
Corynoneura	1	1	4
Cricotopus bicinctus			3
Cricotopus trifascia			3
Cricotopus/Orthocladius	11	5	19
Eukiefferiella	18		6
Forcipomyiinae	1	1	
Hemerodromia	15	3	1
Labrundinia		1	1
Lopescladius	2	1	
Microtendipes	1		
Myxosargus		1	
Parakiefferiella		2	
Parametriochnemus	17	1	
Paratanytarsus			5
Polypedilum aviceps	4		1
Polypedilum convictum	4	1	1
Polypedilum illinoense grp	1		6

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110326], Station #1, Sample Date: 3/24/2011 10:30:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Prosimulium	37		2
Rheocricotopus	22	1	15
Rheotanytarsus	9	1	11
Simulium	26		25
Stempellina		1	
Stempellinella	20	46	6
Tabanus	-99		
Tanytarsus	16	14	10
Thienemanniella	4	2	19
Thienemannimyia grp.	10	17	14
Tipula	-99		
Tvetenia bavarica grp	5		2
EPHEMEROPTERA			
Acerpenna			5
Caenis latipennis	166	100	132
Ephemera simulans		2	
Eurylophella	2		6
Isonychia bicolor	5		
Leptophlebiidae		2	
Maccaffertium mediopunctatum	2		
Maccaffertium pulchellum	45	15	4
Stenacron		3	
Stenonema femoratum	2	6	
ISOPODA			
Caecidotea	3	1	5
Caecidotea (Blind & Unpigmented)		2	
LIMNOPHILA			
Ancylidae		1	
Gyraulus		1	
LUMBRICINA			
Lumbricina		-99	
MEGALOPTERA			
Corydalus	-99		
Nigronia serricornis	1	-99	-99
ODONATA			
Argia	1		
Calopteryx			2
Enallagma			2
Gomphidae	2	1	
Hagenius brevistylus		1	

Aquid Invertebrate Database Bench Sheet Report**Trib. Mineral Fk [110326], Station #1, Sample Date: 3/24/2011 10:30:00 AM****CS = Coarse; NF = Nonflow; RM = Rootmat; -99 = Presence**

ORDER: TAXA	CS	NF	RM
Stylogomphus albistylus			-99
PLECOPTERA			
Acroneuria		-99	-99
Allocapnia	1		
Amphinemura	20		5
Chloroperlidae	3		1
Clioperla clio			-99
Leuctridae	9	15	
Neoperla	8		
TRICHOPTERA			
Cheumatopsyche	10		
Chimarra	10		1
Helicopsyche	1		
Hydropsyche	1		
Hydroptila	4		2
Neureclipsis		1	
Oecetis	1		
Pycnopsyche			1
Rhyacophila	7		
Triaenodes	1		1
TRICLADIDA			
Planariidae	6		1
TUBIFICIDA			
Spirosperma	2		
VENEROIDA			
Pisidiidae			1

Appendix B

Fine Sediment Percent Coverage Statistics: Kruskal –Wallis One Way ANOVA on Ranks with Dunn's Test Multiple Comparisons of Test Stations versus the Control Streams – 2010

Key: **PC** = Pond Creek; **SB** = Shibboleth Branch; **TMF** = Trib. Mineral Fork

One Way Analysis of Variance

Wednesday, September 01, 2010, 11:16:10 AM

Data source: Data 1 in Tribs 2010 Stats

Dependent Variable: Percent Sediment

Normality Test: Failed ($P < 0.050$)

Test execution ended by user request, ANOVA on Ranks begun

Kruskal-Wallis One Way Analysis of Variance on Ranks

Wednesday, September 01, 2010, 11:16:10 AM

Data source: Data 1 in Tribs 2010 Stats

Group	N	Missing	Median	25%	75%
TMF2	18	0	75.000	65.000	97.000
TMF1	18	0	35.000	10.000	75.000
PC 2	18	0	95.000	90.000	95.000
PC 1	18	0	27.000	23.000	55.000
SB3	18	0	15.000	5.000	87.000
SB1	18	0	36.500	15.000	77.000
Control	90	0	7.000	3.000	20.000

$H = 94.789$ with 6 degrees of freedom. ($P = < 0.001$)

The differences in the median values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference ($P = < 0.001$)

To isolate the group or groups that differ from the others use a multiple comparison procedure.

Multiple Comparisons versus Control Group (Dunn's Method) :

Comparison	Diff of Ranks	Q	P<0.05
PC 2 vs Control	118.050	7.979	Yes
TMF2 vs Control	95.467	6.453	Yes
SB1 vs Control	51.828	3.503	Yes
PC 1 vs Control	49.467	3.343	Yes
TMF1 vs Control	44.717	3.022	Yes
SB3 vs Control	37.633	2.544	No

Note: The multiple comparisons on ranks do not include an adjustment for ties.